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Vol: 8 & Iss: 2

Research Article

RF PUBLISHERS

Mosquitoes Composition and *Plasmodium* Infection Status in Malaria Vectors in an Urban Community in Lafia Metropolis, Nasarawa State, Nigeria

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Citation: Ombugadu A, Ladan KH, Aminu R, et al. Mosquitoes Composition and *Plasmodium* Infection Status in Malaria Vectors in an Urban Community in Lafia Metropolis, Nasarawa State, Nigeria. *Int J Cur Res Sci Eng Tech* 2025; 8(2), 309-313. DOI: doi.org/10.30967/IJCRSET/Akwashiki-Ombugadu/184

Received: 15 May, 2025; Accepted: 20 May, 2025; Published: 22 May, 2025

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ABSTRACT

Background: Malaria continues to be a significant threat to public health in Nigeria and the best way to nip it on board is through entomological surveillance. Therefore, this study aimed at investigating mosquitoes composition and *Plasmodium* infection status in malaria vectors in an urban community in Lafia metropolis of Nasarawa State, Nigeria.

Method: The survey was carried out during wet season between June and July 2021. Prokopack Aspirator was used to collect indoor resting mosquitoes from 6:00 am to 9:00 am from 30 randomly selected houses. Mosquitoes were sorted and identified using standard identification keys. Malaria vectors were dissected and screened for the presence of sporozoite.

Results: A total of 161 mosquitoes were collected which spread across three mosquito species namely, *Culex quinquefasciatus* 84 (52.17%), *Anopheles gambiae s. l.* 69 (42.86%) and *Aedes aegypti* 8 (4.97%). The abundance of mosquitoes in relation to species varied significantly (P < 0.05). Most of the inhabitants in the randomly selected households use insecticides in their rooms while some other households sleep under long lasting insecticidal treated bed nets. The abundance of mosquitoes in the month of June was low 70 (43.48%) while July period recorded higher number of mosquitoes 91 (56.52%). Fed female mosquitoes were the most dominant 53 (48.18%) followed by unfed 36 (32.73%) then gravid 12 (10.91%) while the least was half gravid females 9 (8.18%). None of the 48 female *An. gambiae s. l.* dissected had the infective stage of *P. falciparum*. The entomological indices in the study area shows a 3.66 mosquitoes per house per night and an overall 1.18 bites/person/night.

Conclusion: In conclusion, the inhabitants of the area should always sleep under insecticide treated bed nets and make use of other relevant forms of protection against mosquitoes bites.

Keywords: Mosquitoes; Vectors; Prokopack Aspirator; Entomological transmission indices; Wet season

Introduction

The impact of climate change is thwarting malaria eradication efforts; hence, this nexus should be looked into very critically¹ most especially for the fact that urban centers anthropogenic activities greatly contribute to carbon emissions to a large extent. Malaria remains a significant public health concern worldwide affecting livelihood and human wellbeing². Africa in particular bears the greatest burden of malaria, accounting for nearly 90% of malaria cases in Africa and Nigeria bear a significant amount of the numbers³. Malaria is mainly transmitted by the different species of mosquitoes which are responsible continuous spread of the disease⁴. Malaria is transmitted throughout Nigeria, approximately 97% of the population at risk of malaria infection⁵. Plasmodium falciparum is the predominant malaria species while the primary vector across the country is Anopheles (An.) gambiae s. s., accounting for 67.1%, with An. funestus as a secondary vector in some areas of Nigeria⁵. According to the 2021 World Malaria Report, Nigeria had the highest number of global malaria cases (27 % of global malaria cases) and the highest number of deaths (32 % of global malaria deaths) in 2020⁶ and accounted for an estimated 55.2% of malaria cases in West Africa in 2020.

Efforts to control malaria relies chemotherapeutic and vector control approaches which remains bedrock of malaria elimination strategies7. The use of WHO approved artemisininbased combination therapy (ACT) have achieved substantial success in the past. Sadly, the ambitious goal of malaria elimination and eradication agenda is far from reality due to drug resistance to the mainstream treatment options while vector control is being adopted as key component of for transmission blocking via the use of insecticide treated nets, indoor residual spray⁶. As such vector control remains the frontline strategy for preventing and reducing malaria transmission⁸. Implementation of cost-effective vector control interventions rely on adequate knowledge of local vector species attained through sustained entomological monitoring and surveillance activities9. Such surveillance activities provide information on the dynamics of vector species composition, relative abundance and sporozoites infection rates¹⁰.

Besides, these and other entomological indices such as vector behavior and insecticide susceptibility status also serve as the basis for measuring the impact of vector control strategies on malaria transmission⁹. Significant spatial heterogeneities have been observed in the abundance of indoor resting *Anopheles* mosquitoes, their species composition and sporozoite infection rates¹¹. Characterization of entomological indices and mosquito behavior in different settings, therefore, becomes necessary to guide the suitability of interventions such as long-lasting insecticide nets (LLINs) and indoor residual spray (IRS) designed to target indoor biting or resting mosquitoes¹¹.

Notable reductions in malaria burden have been observed across the globe in past decade but the recent emergence of the coronavirus pandemic has significantly impeded these gains¹². Thus, the main priority is to reduce global mortality and morbidity and to reduce or eliminate the vector population¹³. Malaria surveillance at community level is important, as this has the potential to give unbiased indication of geographical variation and trends over time, but such data are lacking from most areas so that trends are only estimated by modelling¹⁴. Separate small-scale surveys conducted in some communities in Nigeria over the past decade have indicated continuing high levels of malaria parasite infection, particularly in areas of the north with prevalence still exceeding 60%¹⁴. Here, we analyzed the composition of mosquito species and malaria transmission within a community in Lafia metropolis, Nasarawa State, Nigeria. Emerging from this analysis are details of trends of malaria vector composition that are informative for formulating future intervention strategies.

Materials And Methods

Study area

The study was conducted in Kofar-Kaura community with the coordinates 8° 48'98"N and 8° 51'36"E in Lafia Local Government Area (LGA) of Nasarawa State, Nigeria. Lafia Local Government Area has a population of 330,712 inhabitants based on the National Population Census of 2006. Rainfall is moderately high in Lafia, ranging from 1200 mm to 1600 mm¹⁵. Average maximum and minimum daily temperatures are 35°C and 21°C in rainy season and 37°C and 16°C in dry season, respectively¹⁶. The study site is an urban settlement with agricultural activities taking place in some part of the settlement.

Ethical consent

The Chief, elders and head of households in Kofar-Kaura community were consulted and informed on the significance of the research to the community. The Chief granted permission for the research work to be carried out by endorsing the ethical consent form. Also, all the head of households consented too.

Mosquitoes sampling: Prokopack aspirator collection

The study lasted for a two-month period, from June to July, 2021. Thirty houses were randomly selected for the purpose of this study. The head of households were contacted and informed on the eve to collection day in order for them to ensure that doors and windows are kept close during the morning hours when they were awake until the rooms were sampled. Selected houses were revisited twice throughout the periods of the study. The Prokopack Aspirator which is being powered by a battery is simple, maneuverable, less laborious, less time consuming and eco-friendly device according to Ombugadu, et al¹⁷, was used for sampling of mosquitoes early in the morning between 0600 to 0900 hours for three days (i.e. ten households per day) in each of the two entomological surveillance months. The Prokopack Aspirator was turned on and off with a rotary switch located at the handle. After each household's collection was made, the lid on the collection cup was covered properly before turning off the unit. Thereafter, moist cotton wool that was soaked in chloroform was used to knockdown mosquitoes collected for a period of five minutes, after which samples were transferred into a well-labeled petri dish and transported to the Department of Zoology Laboratory of Federal University of Lafia, Nasarawa State, Nigeria for sorting, morphological identification and further processing.

Morphological identification

Collected mosquito species in petri dishes were morphologically identified using a dissecting microscope and identification keys by Gillies and Coetzee¹⁸ and Kent¹⁹, respectively.

Screening of malaria vectors for sporozoite

All *Anopheles* mosquitoes collected were dissected using a dissecting stereomicroscope and sterilized dissecting needles

in order to detect the Plasmodium parasites infective stage as described by Centers for Disease Control and Prevention²⁰ and WHO²¹.

Statistical analysis

Data obtained was analyzed using R Console software (version 3.2.2). Pearson's Chi-square test was used to compare mosquitoes' proportions in relation to months, species, sex and abdominal conditions, respectively. The P-value < 0.05 was considered statistically significant.

Entomological transmission indices

The sporozoite rate, indoor resting density and man biting rate indices were measured as described by William and Pinto²².

Sporozoite rate (S): The number of female anopheline mosquitoes found with sporozoite after their salivary glands dissection was divided by the number of female anopheline dissected as described by Williams and Pinto²² in equation 1.

 $S = \frac{\text{Number of sporozoites positive mosquitoes}}{\text{Number of mosquitoes dissected}} \dots (1)$

Indoor resting density (IRD) of female mosquitoes

The indoor resting density of female *Anopheles* per structure per night was calculated using the formula by Williams and Pinto²² as expressed in equation 2.

 $IRD = \frac{\frac{\text{Total number of female vectors collected}}{\text{Total number of houses sampled}} \dots (2)$

Man biting rate (MBR) of female mosquitoes

All the females (F) collected were separated and counted in relation to species and sexes that they belong to. The total number of collected females of each species was divided by the total number of occupants (W) who spent the previous night in the rooms sampled as shown in equation 3²².

$$MBR = \frac{Female mosquitoes collected (F)}{Total number of occupants (W) in the households sampled} \dots (3)$$

Results

Composition and abundance of mosquito species in kofarkaura community

Mosquito species checklist generated at the end of this study is shown in **Table 1**. The overall total of 161 mosquitoes were caught resting indoors. The 161 mosquitoes spread across a family, two subfamilies, three genera and three species. The highest number of resting indoor mosquitoes was *Culex quinquefasciatus* 84 (52.17%), followed by *Anopheles gambiae* 69 (42.86%) and the least was *Aedes aegypti* 8 (4.97%). The abundance of each mosquito species between the months showed no significant difference (*Ae. aegypti*: $\chi^2 = 0.14694$, df = 1, P = 0.7015; *An. gambiae*: $\chi^2 = 0.29985$, df =1, P = 0.584; *Cx. quinquefasciatus*: $\chi^2 = 0.37149$, df =1, P = 0.5422).

Table 1: Checklist of Mosquito Species in Kofar-KauraCommunity, Lafia Metropolis, Nasarawa State, Nigeria.

Species	Mosquito Abur	Total (%)	
	June	July	
Aedes aegypti	3 (4.30)	5 (5.50)	8 (4.97)
Anopheles gambiae	28 (40.00)	41 (45.05)	69 (42.86)
Culex quinquefasciatus	39 (55.70)	45 (49.45)	84 (52.17)
Total (%)	70 (43.48)	91 (56.52)	161

Sex-wise abundance of mosquitoes

Table 2 shows the abundance of mosquitoes in relation to sex. The female indoor resting mosquitoes were higher 110 (68.32%) than the males 51 (31.68%). The abundance of mosquitoes for each species in relation to sex showed a high significant difference (*Ae. aegypti*: $\chi^2 = 25$, df =1, P = 5.733e⁻⁰⁷; *An. gambiae*: $\chi^2 = 15.319$, df =1, P = 9.078e⁻⁰⁵; *Cx. quinquefasciatus*: $\chi^2 = 11.116$, df =1, P = 0.0008561).

Table 2: Abundance of Mc	squitoes in	Relation to	Sex
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Species	Sex	Total (%)	
	Male (%)	Female (%)	
Ae. aegypti	2 (25.00)	6 (75.0)	8 (4.97)
An. gambiae	21 (30.43)	48 (69.57)	69 (42.86)
Cx. quinquefasciatus	28 (33.33)	56 (66.67)	84 (52.17)
Total (%)	51 (31.68)	110 (68.32)	161

Abdominal conditions of female mosquitoes

The abundance of mosquitoes in relation to abdominal condition is shown in Table 3. Most of the female mosquitoes were fed 53 (49.10%) followed by unfed 47 (29.20%), half gravid 19 (11.80%) and the least was gravid 16 (9.90%) as shown in Table 3. The overall abdominal conditions varied significantly (χ^2 = 39.19, df = 3, P = 1.109 e⁻⁰⁸). Also, the abdominal conditions for each species varied significantly (P < 0.05), respectively.

Table 3: Abundance of Mosquitoes in Relation to Abdominal Conditions.

Species	Abdominal Conditions (%)				Total (%)
	Unfed	Fed	Half gravid	Gravid	
Aedes aegypti	3 (50.00)	1 (16.67)	2 (33.33)	0 (0.0)	6 (5.45)
Anopheles gambiae	12 (25.00)	28 (58.33)	3 (6.25)	5 (10.42)	48 (43.64)
Culex quinquefasciatus	21 (37.50)	24 (42.86)	4 (7.14)	7 (12.5)	56 (50.91)
Total (%)	36 (32.73)	53 (48.18)	9 (8.18)	12 (10.91)	110

Prevalence of the infective stage malaria parasite in the vector

The prevalence of malaria parasite in the study area showed no positive result. Sporozoite was absent in all the 48 mosquitoes dissected as shown in **Table 4**.

Table 4: Prevalence of Sporozoites in Anopheles gambiae in two months period.

Month	No. of An. gambiae dissected	No. positive for sporozoites (%)
June	21	0 (0.00%)
July	27	0 (0.00%)

Malaria entomological transmission indices

The entomological transmission indices of mosquitoes in Kofar-Kaura Community as shown in **Table 5** revealed that the overall indoor resting density was 3.66 *Anopheles* mosquito/house/night while the pooled man biting rate for the two months surveyed was 1.18 *Anopheles* mosquito bite/man/ night. Although the indoor resting density of female mosquitoes per household was higher in the month of July 2.03 *Anopheles* mosquito/house/night than in June 1.63 *Anopheles* mosquito/ house/night. Also, man biting rate per human in the households was high in the month of July 0.66 mosquito/man/night than in June 0.52 mosquito/man/night.

Table 5: Entomological Transmission Indices of Mosquitoes in Kofar-Kaura Community.

Index	Months		Overall
	June	July	
Indoor resting density	1.63	2.03	3.66
Man biting rate	0.52	0.66	1.18

Discussion

The abundant and diverse mosquito species recorded in the study area falls within a taxonomic family, two sub-families, three genera and three species. The study site may likely favor the breeding of mosquito species and may result in rapid transmission of mosquito-borne disease through bite by infected mosquito species within Kofar-Kaura community. This study is similar to the findings of Ebenezer et al.²³ who recorded a family, two sub-families, three genera and five species in a study of spatial distributions and indoor resting density of mosquito species in lowland rainforest of Bayelsa State, Nigeria.

The observed variation of mosquito's population in favor of the month of July having 91 individuals (56.52%) could be due to high amount of rainfall that gave rise to more breeding sites and higher relative humidity. This is in disagreements with the findings of Lamidi²⁴ in a study of distribution and seasonal abundance of *Anopheles* mosquito species in Nguru, Yobe State, Nigeria who stated that the abundance of *Anopheles gambiae* rises in August and fell in September.

The sex-wise preponderance of female mosquitoes to males in this study maybe genetically driven. This is similar to the finding of Landscape²⁵ whose 4,477 trapped mosquitoes had dominant number of female mosquitoes 2345 (52.40%) over males 2132 (47.60%).

The blood-fed females accounting for the abdominal condition with greatest proportion, possibly suggests high human-vector contact in the study area thereby making them highly/very vulnerable to mosquito-borne infections if bitten by any infected female mosquito. This is in disagreements with the findings of Ebenezer et al.²³ who showed that unfed female mosquitoes were the highest 2098 (45.90%) followed by fed 1888 (41.30%) then gravid 478 (10.50%) and the least was half gravid 102 (2.20%).

The absence of *Plasmodium* infective stage (sporozoite) in the screened mosquitoes as at the time of this study possibly implies that the trapped mosquitoes are either nulliparous or have not fed on infected individuals. Also, it shows that the prevalence malaria in the study area maybe low. This agrees with other studies by The PMI Vector Link Project^{26,27}, Abdelwhab et al.²⁸ and Osidoma et al.²⁹ who recorded a zero percent (0.0%)prevalence of sporozoite in Anopheles mosquitoes. Also, Aju-Ameh et al.³⁰ recorded a sporozoite rate of 0.0% in urban area of Benue State. On the contrary, Njila et al.³¹ reported a sporozoite rate of 66.7% in a study on infection status of mosquitoes in a community in Jos North LGA of Plateau State, Nigeria. Also, the findings of Bigoga et al.³² stated that of the 1179 mosquitoes examined for P. falciparum circumsporozoite antigen by ELISA across the months of both seasons 103 (8.76%) mosquitoes had the infective stage of the malaria parasite.

The pooled indoor resting density of female mosquito species across the months in this study was 3.66 mosquitoes/room/night which is in accordance with the findings of Ombugadu et al.¹⁷ who recorded an indoor resting density of female mosquitoes to be 1.625/room/night. Similarly, a study conducted by Ebenezer et al.²³ on indoor resting density of female mosquito species in Bayelsa State recorded 2.9-3.8 mosquitoes/room/night.

The overall man biting rate of female mosquito species across the months of this study of 1.18 bites/person/night. This is in line with the previous finding by Ombugadu et al.¹⁷ who reported an overall man biting rate of female mosquito species to be 0.51 bite/person/night. Likewise, our finding is not similar to the work by Ebenezer et al.²³ who recorded 19.10 bites/person/ night in Bayelsa State.

Conclusion

This study shows that Kofar-Kaura community is a breeding site for mosquito vectors which can possibly transmit mosquito-borne infections. The population of mosquitoes in the area across the months was dynamic. Most of the *Anopheles* mosquitoes collected were blood fed which implies high human-vectors contact. *Plasmodium* infective stage was not found in the screened malaria vector. Conclusively, the inhabitants of the area should control mosquito-borne infections through clearing of stagnant water bodies around their environment and judiciously make use of insecticide treated bed nets. Also, the time spent outdoors for nocturnal activities by the people in the area should reduce in order for them to avoid any possible outdoor transmission of malaria infection.

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