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Full Length Research Article

FEEDING AND RESTING PATTERN OF MALARIA VECTORS IN THREE COMMUNITIES FROM SOUTHEAST NIGERIA

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ABSTRACT

Accurate knowledge of the feeding and resting time of malaria vectors is very crucial for malaria management, hence a 12-month longitudinal study was done to tackle the challenge of transmission of mosquito-borne diseases. Centre for Disease Control (CDC) light traps and the Human Landing Catch (HLC) methods were used to sample indoor and outdoor malaria vectors at Nibo, Mgbakwu and Enugwu-Agidi communities in Anambra State, Nigeria. Chi-square test was used to check for significant difference ($P<0.05$) between feeding and resting time and other parameters in the study. Result showed that malaria vectors were caught both outdoors and indoors which indicated that they have a crepuscular and concurrent resting/feeding time between 6 pm-12 am and 12 am - 6 am. This implies that the malaria vectors were non-specific in their feeding time, recording high peak feeding time of 24.4%, 18.3% and 17.9% between 6am - 12pm and 1am – 6am at Nibo, Mgbakwu and Enugwu-Agidi respectively. This heterogenic nature and behavior of malaria vectors may have caused a lot of hindrances to malaria control strategies and interventions deployed in the past. Therefore, proper understanding of behaviours of malaria vectors will help in evidence-based policy decision for strategic malaria control in Anambra State.

Keywords: Malaria vector, feeding and resting pattern, Anambra State

INTRODUCTION

Mosquitoes which belong to the family culicidae causes nuisance and also a major public health problem because the females feed on blood of vertebrates, including humans, and in doing so transmit harmful diseases such as malaria, yellow fever and filariasis (Obol *et al*., 2021). They are among the major causes of illness to more than 700 million people annually and responsible for the death of about 1 in 17 particularly in the tropical and subtropical countries of the world (WHO, 2020a). In Nigeria, Anopheline vectors are known for their biting annoyance, noise nuisance, sleeplessness and allergic reaction from their bites (Kumar *et al.*, 2014). Members

of the *An. gambiae* complex are important malaria vectors in sub-Saharan Africa but differ strongly in their host-preference which is assumed to be stimulated by the hosts' odour (Gatton *et al.,* 2013). *Anopheles arabiensis* occupies over 70% of sub-Saharan Africa, dominates in arid zones and some of the highland areas. It adapts to endophagic and endophilic patterns where hosts are domestic and indoor but adopts exophagic patterns where hosts are mainly outdoors (Sherrard-Smith *et al*., 2019).

Sleeping under insecticide-treated bed nets could reduce malaria transmission by limiting contact between mosquito vectors and human hosts. However, malaria vectors can also feed at dusk and dawn when people are not protected. In addition, there are growing reports of behavioural resistance, such as changes in mosquito biting behaviour (i.e. "anti-insecticide" behaviour), which reduce the probability of insecticide encounter which equally attenuate the efficacy of insecticides (Tiono *et al*., 2018). In some countries in Africa especially Nigeria, the principal vectors of malaria have shown consistent biting patterns and remain amenable to the effective Indoor Residual Spray and Long Lasting Insecticide Treated Nets interventions (Gerald and Larry, 2010), whereas in other places, high levels of outdoor biting by *An. gambiae* sensu stricto was observed throughout the night, including during early evening and morning hours when human hosts are often outdoors (Cheikh *et al*., 2015). Furthermore, *An. funestus* has shown a behavioural change in biting activity where it remained both anthropophilic and endophilic, while adopting diurnal feeding when local people are not protected by IRS and LLINs (Chouibon *et al*., 2016) but become completely exophilic in response to indoor spraying. However, following the adoption and scale-up of IRS and LLINs, a shift in mosquito behaviors has been observed, where mosquitoes more often bite humans outdoors, and earlier in the evening, thereby avoiding insecticide treated surfaces and thereby threatening the effectiveness of the interventions (Killeen, 2014 and Chow *et al*., 1993). These situations are alerts that the surveillance and monitoring of malaria and its vectors should be both regular and consistent (Coetzee *et al*., 2019).

To incriminate the vectors and understand the transmission dynamics of malaria in an endemic area like Anambra State, studies on the entomological parameters like the human biting rate, sporozoite rate, blood meal source, human blood index, entomological inoculation rate, feeding patterns and resting behaviour of malaria vectors are crucial. This study explored feeding time of malaria vectors in three communities of Nibo, Mgbakwu, and Enugwu-Agidi in Anambra State, south east Nigeria. Findings from this study will help to better understand the potential epidemiological consequences of shifts in feeding time and resting behaviour of malaria vectors.

MATERIALS AND METHODS

Study area

The study was carried out between September, 2021 and August, 2022 in Ezeawulu and Umuanum villages of Nibo in Awka South L.G.A., Amaezike and Akamanator villages of Mgbakwu in Awka North L.G.A., Umu Agidi and Ifite villages of Enugwu Agidi in Njikoka L.G.A. of Anambra Central Senatorial Zone of Anambra State, Southeast Nigeria. Anambra Central Senatorial district is located between Lat. 6˚ 12'45.68''N to Lat. 6˚ 22'45.68''N and Long. 7° 04'19.16''E to Long. 7° 15'19.16''E of Greenwich with an average daily temperature of 26.8 °C / 80.2 °F (Microsoft Encarta, 2009). The geographical coordinates of Awka South LGA is located between Lat. 6˚10' North to Lat. 6˚23' N of equator and Long. 7° 04' E to 7° 44' E of Greenwich, Awka North is located between Lat. 6° 15' N to

6° 35' N of equator and Long. 7°10' E to 7°30' E of Greenwich while Njikoka LGA is lying between Lat. 06° 20' 58 '' N to 06° 21' 00'' N and Long. 06° 12' 55''E to 06° 52' 55''E of Greenwich. The study area has two major seasons, the rainy season (April to October) and the dry season (November to March), with a spell of harmattan - a period of cold weather with misty atmosphere (Enete, 2008) - between November and January. The total annual rainfall of the State is above 11,450mm for the six to seven months of the rainy season whereas the mean annual temperature range is 30.0 – 36.0ºC (Enete, 2004). The official language of the people is Igbo although English language is widely spoken throughout the state as a secondary language (Microsoft Encarta, 2009). The villages are an agricultural trade centre where yams, cassava, corn (maize), palm oil and kernels are sold for to the people of the surrounding area. (Microsoft Encarta, 2009). The environmental problems and lack of proper sanitation encourages the breeding of malaria vectors and malaria transmission in these communities (Figure 1).

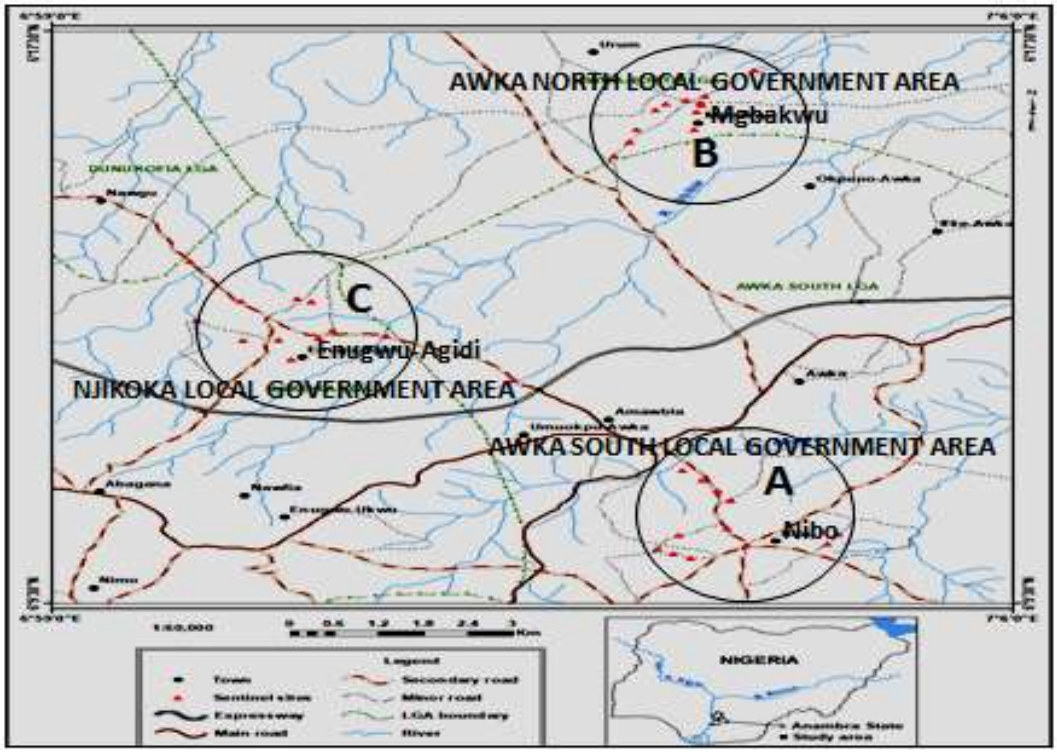


Figure 1: Map of study area showing Nibo [A], Mgbakwu [B] and Enugwu-Agidi [C]. Anambra State, Nigeria (in-set). Adapted from Google Map 20th December, 2022)

Study design

This was a longitudinal study design in which mosquito vectors were captured monthly using CDC light trap and Human Landing Catch methods to determine their feeding and resting times during the wet and dry seasons, between September 2021 and August 2022.

Selection of villages and households for mosquito sampling:

One village was randomly selected from all villages that make-up the respective target communities. Different sites were again randomly selected from each village. According to the method described by WHO (1992), households for mosquito sampling were randomly selected from a pool of houses from the selected sites. Each selected village had equal representation in terms of numbers of sites and households studied.

Advocacy and Community Mobilization

A letter of introduction was obtained from the Department of Parasitology and Entomology, Nnamdi Azikiwe University, Awka Anambra State seeking for cooperation from the study participants to carry out the research in the communities of Nibo in Awka South LGA, Mgbakwu in Awka North LGA and Enugwu-Agidi in Njikoka LGA of Anambra State southeast, Nigeria. Three levels of advocacy were carried out; first to the heads in charge of Health Department through the Local Government Chairman of the selected LGAs, followed by advocacy visit to the community leaders/president general of the selected LGAs and finally to the households/individuals of the selected communities that will participate in the study. Approval for study participation was obtained before the commencement of the study and the identity of the participants were anonymous during and after the survey.

Sampling procedures for entomological investigations

The CDC light traps and human landing catches were the two methods used for collection of outdoor and indoor mosquito vectors in the study.

Collection of adult mosquito vector

The collections were performed monthly during three consecutive nights/days, and each collection period lasted for twelve hours, from 6pm to 6am for the Human Landing Catch (HLC) and CDC Light Trap. The CDC light traps were placed approximately 1.2m above the floor in and outside the room as described by WHO (2015). The CDC miniature light trap uses light as an attractant to lure mosquitoes into the collection chamber. The trap contains an impeller fan designed to capture and retain the mosquitoes inside the specimen cups. Traps were switched on from 6 pm and removed by 6 am the next day. The CDC miniature light trap uses light as an attractant to lure mosquitoes into the collection chamber. The trap contains an impeller fan designed to capture and retain the mosquitoes inside the specimen cups. Each night, 6 traps (one trap indoor and one trap outdoor at three different locations) were set for three nights for each site each month and they were rotated in the same order each month. The HLC was performed for 50 minutes each hour with 10 minutes break for the collectors. At each house two adults were stationed outdoors 10m from the house, and two were stationed indoors. Outdoor collections were conducted from 6.00pm to 9.50pm, after which time few people were outdoors, and indoor collections from 6.00pm to 5.50am. Collectors sat on chairs with their legs exposed. Using flashlights, collectors caught landing mosquitoes with a hand – held mouth aspirator and test tubes and each hour's collection were kept separately; and the collectors were rotated between sites on different nights.

All adult mosquitoes collected were temporary preserved on moist filter papers in petri dishes as described by WHO (2015) and transferred to the Vector Research Department at the National Arbovirus and Vectors Research Centre (NAVRC), Enugu, Nigeria for identification.

Morphological identification of malaria vectors

All the mosquito vectors collected were identified and sorted out under a Stereomicroscope Leica model NSW series IMNS 210 entomological microscope. The mosquitoes were sorted out and malaria vectors were subsequently identified using morphological keys of Gillies and De Meillion (1968); Gillies and Coetzee (1987).

Analysis of Data

Data obtained from the study were summarized and analyzed using descriptive and comparative statistics. Mean and standard deviation, one-way analysis of variance (ANOVA) and the least significant difference (LSD) test were used for multiple comparison of feeding and resting pattern of malaria vectors.

Results:

Feeding time of malaria vectors caught with CDC light traps at Nibo, Awka South LGA, Anambra State

The feeding time of malaria vectors caught with CDC light traps at Nibo showed that 25.8% of malaria vectors fed/rested between 6-7pm followed by 17.9% collected between 11pm-12am whereas 16.6% of malaria vectors fed/rested between 3-4am and 4-5am respectively as shown in Table 1.

Table 1: Feeding time of malaria vectors caught with CDC light traps at Nibo, Awka South LGA, Anambra State

Feeding Time	Numbers of mosquitoes caught in three consecutive dusk to dawn (Sept. 2021- Aug. 2022)													
	Sep	Oct	Nov	Dec	Jan	Feb	Mar	Apr	May	Jun	July	Aug	Total	Rel.%
6-7p.m	3	0	0	1	4	1	12	1	3	2	10	2	39	25.8
7-8	0	0	0	0	0	0	0	0	0	0	0	0	0	0.0
8-9	0	0	0	0	0	0	0	0	0	0	0	0	0	0.0
9-10	0	0	0	2	1	0	5	1	1	0	5	0	15	9.9
10-11	0	0	0	0	0	0	0	0	0	0	0	0	0	0.0
11-12	0	0	0	0	2	1	5	0	2	3	10	4	27	17.9
12-1	2	0	0	0	0	2	0	0	0	2	0	0	6	3.9
1-2	1	0	0	1	0	0	0	0	1	0	2	0	5	3.3
2-3	1	0	0	0	2	2	1	0	0	1	1	0	8	5.3
3-4	0	0	0	1	2	4	4	0	0	2	12	0	25	16.6
4-5	0	0	0	0	0	1	2	1	0	0	15	6	25	16.6
5-6	0	0	0	0	0	1	0	0	0	0	0	0	1	0.7
Total	7	0	0	5	11	12	29	3	7	10	55	12	151	100
Relative %	4.6	0.0	0.0	3.3	7.3	8.0	19.2	2.0	4.6	6.6	36.4	8.0	100	

***Anopheles* hourly aggressiveness from 6 pm to 6am. CDC: Centre for Disease Control Light Trap Collection; VFT: Vector Feeding Time 6–11: p.m.; 12–6: am; –: number of mosquito collected.**

Feeding time of malaria vectors caught with CDC light traps at Mgbakwu, Awka North LGA, Anambra State

The feeding time of malaria vectors caught with CDC light traps at Mgbakwu showed that 26.0% of malaria vectors fed/rested between 8-9pm followed by 24.4% collected between 6pm-7pm whereas 15.8% of malaria vectors fed/rested between 9-10pm as shown in Table 2.

Table 2: Feeding time of malaria vectors caught with CDC light traps at Mgbakwu, Awka North LGA., Anambra State

Time	Numbers of mosquitoes caught in three consecutive dusk to dawn (Sept. 2021- Aug. 2022)													
	Sep	Oct	Nov	Dec	Jan	Feb	Mar	Apr	May	Jun	July	Aug	Total	Rel.%
6-7p.m	3	0	2	0	1	4	0	0	4	10	7	0	31	24.4
7-8	1	0	1	0	0	2	2	0	3	0	1	1	11	8.7
8-9	1	1	2	0	0	5	3	0	7	10	4	0	33	26.0
9-10	2	1	1	0	0	1	7	0	0	0	8	0	20	15.8
10-11	0	0	0	0	0	0	0	0	0	0	0	0	0	0.0
11-12	1	0	1	0	0	0	1	2	0	0	1	2	8	6.3
12-1a.m	0	0	0	0	0	0	0	0	0	0	0	0	0	0.0
1-2	1	1	1	0	0	0	1	1	0	0	0	0	5	3.9
2-3	1	2	1	0	0	0	0	0	0	0	2	0	6	4.7
3-4	0	0	1	0	0	0	0	1	2	0	1	1	6	4.7
4-5	0	0	0	0	0	0	0	0	0	0	0	0	0	0.0
5-6	2	0	0	0	1	0	3	0	0	1	0	0	7	5.5
Total	12	5	10	0	2	12	17	4	16	21	24	4	127	100
Relative %	9.4	3.9	7.9	0.0	1.6	9.4	13.4	3.2	12.6	16.5	18.9	3.2	100	

***Anopheles* hourly aggressiveness from 6 pm to 6am. CDC: Centre for Disease Control Light Trap Collection; VFT: Vector Feeding Time 6–11: p.m.; 00–6: am; –: number of mosquito collected.**

Feeding time of malaria vectors caught with CDC light traps at Enugwu-Agidi, Njikoka LGA, Anambra State

The feeding time of malaria vectors caught with CDC light traps at Enugwu-Agidi showed that 20.7% of malaria vectors fed/rested between 9-10pm followed by 17.2% collected between 7pm-8pm whereas 13.6% of malaria vectors fed/rested between 8-9pm and 11pm-12am respectively as shown in Table 3.

Table 3: Feeding time of malaria vectors caught with CDC light traps at Enugwu-Agidi

	Numbers of mosquitoes caught in three consecutive dusk to dawn (Sept. 2021- Aug. 2022)													
Time	Sep	Oct	Nov	Dec	Jan	Feb	Mar	Apr	May	Jun	July	Aug	Total	Rel.%
6-7p.m	0	0	0	0	0	0	0	0	0	0	0	0	0	0.0
7-8	7	0	0	4	0	1	0	5	0	2	10	0	29	17.2
8-9	6	0	0	4	0	1	0	5	1	0	6	0	23	13.6
9-10	10	0	0	6	0	1	0	0	1	1	16	0	35	20.7
10-11	0	0	0	0	0	0	0	0	0	0	0	0	0	0.0
11-12	5	0	0	0	0	1	0	0	1	5	11	0	23	13.6
12-1a.m	3	0	0	0	0	0	0	0	0	0	3	0	6	3.5
1-2	5	0	0	2	0	1	1	0	0	4	8	0	21	12.4
2-3	3	0	0	0	0	0	0	5	0	0	2	0	10	5.9
3-4	0	0	0	0	0	0	0	0	0	0	5	0	5	3.0
4-5	0	0	0	0	0	0	0	0	0	0	0	0	0	0.0
5-6	2	0	0	3	0	3	3	5	0	0	0	1	17	10.1
Total	41	0	0	19	0	8	4	20	3	12	61	1	169	100
Relative %	24.3	0.0	0.0	11.2	0.0	4.7	2.4	11.8	1.8	7.1	36.1	0.6	100	

***Anopheles* hourly aggressiveness from 6 pm to 6am. CDC: Centre for Disease Control Light Trap Collection; VFT: Vector Feeding Time 6–11: p.m.; 00–6: am; –: number of mosquito collected.**

Comparison between collections of malaria vectors using CDC light traps and HLC sampling methods

The comparative result between the two collection methods showed that the relative percentage of malaria vectors caught using CDC Light Trap method where high between 6pm and 12am when compared with the relative percentage of malaria vectors caught between 12am and 6am using Human Landing Catch (HLC) method in the hourly collections as shown in Figure 2.

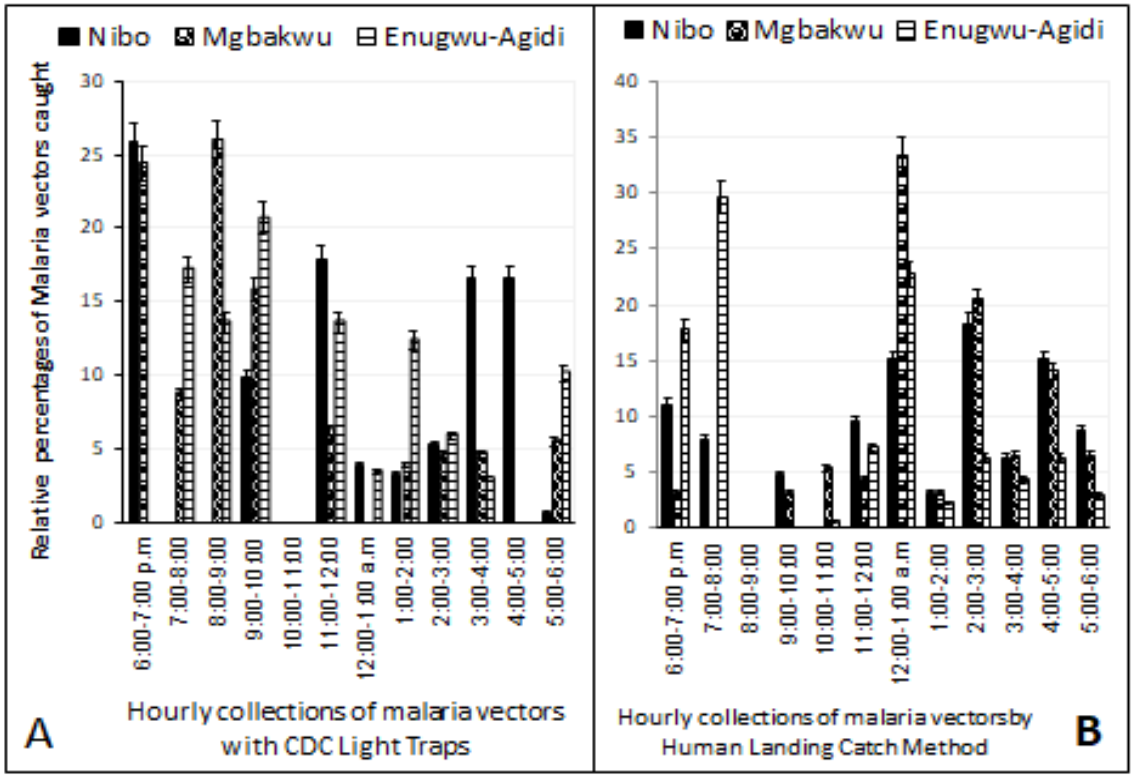


Figure 2: Comparison of collections of malaria vectors using CDC light traps [A], and human landing catches [B] in three communities from Anambra State, Nigeria

Feeding time of malaria vectors caught by human landing catches at Nibo, Awka South LGA, Anambra State

The feeding time of malaria vectors caught with HLC traps at Nibo showed that 18.3% of malaria vectors fed/rested between 2-3am followed by 15.1% collected between 12-1am and 4-5am respectively whereas 11.1% of malaria vectors fed/rested between 6-7pm as shown in Table 4.

Table 4: Feeding time of malaria vectors caught by human landing catches at Nibo, Awka South LGA, Anambra State

Time	Numbers of mosquitoes caught from three consecutive dusk to dawn (Sept. 2021- Aug. 2022)													
	Sep	Oct	Nov	Dec	Jan	Feb	Mar	Apr	May	Jun	July	Aug	Total	Rel.%
6-7p.m	2	4	0	3	0	0	1	3	0	0	1	0	14	11.1
7-8	1	0	0	3	0	0	0	2	0	0	1	3	10	7.9
8-9	0	0	0	0	0	0	0	0	0	0	0	0	0	0.0
9-10	0	0	0	3	0	1	1	0	0	0	0	1	6	4.8
10-11	0	0	0	0	0	0	0	0	0	0	0	0	0	0.0
11-12	0	2	0	3	1	4	2	0	0	0	0	0	12	9.5
12-1a.m	3	2	0	5	1	1	0	0	0	0	4	3	19	15.1
1-2	1	2	0	0	0	0	0	1	0	0	0	0	4	3.2
2-3	3	1	0	1	0	0	5	5	0	0	4	4	23	18.3
3-4	2	0	0	0	0	4	1	1	0	0	0	0	8	6.3
4-5	3	4	0	0	0	1	1	5	0	0	3	2	19	15.1
5-6	1	0	0	0	4	3	0	0	0	0	3	0	11	8.7
Total	16	15	0	18	6	14	11	17	0	0	16	13	126	100
Relative %	12.7	11.9	0.0	14.3	4.8	11.1	8.7	13.5	0.0	0.0	12.7	10.3	100	

***Anopheles* hourly aggressiveness from 6 pm to 6am. Human Landing Catch Collection Method (HLC). VFT: Vector Feeding Time; 6–11: p.m.; 00–6: am; –: number of mosquito collected.**

Feeding-time of malaria vectors caught by human landing catches at Mgbakwu, Awka North LGA, Anambra State

The feeding time of malaria vectors caught with HLC traps at Mgbakwu showed that 33.3% of malaria vectors fed/rested between 12-1am followed by 20.4% collected between 2-3am whereas 14.0% of malaria vectors fed/rested between 4-5am as shown in Table 5.

Table 5: Feeding-time of malaria vectors caught by human landing catches at Mgbakwu, Awka North LGA, Anambra State

Time	Numbers of malaria vectors caught in three consecutive dusk to dawn (Sept. 2021- Aug. 2022)													
	Sep	Oct	Nov	Dec	Jan	Feb	Mar	Apr	May	Jun	July	Aug	Total	Rel.%
6-7p.m	0	3	0	0	0	0	0	0	0	0	0	0	3	3.2
7-8	0	0	0	0	0	0	0	0	0	0	0	0	0	0.0
8-9	0	0	0	0	0	0	0	0	0	0	0	0	0	0.0
9-10	1	0	0	0	0	0	0	0	0	2	0	0	3	3.2
10-11	2	1	2	0	0	0	0	0	0	0	0	0	5	5.4
11-12	0	3	0	0	0	0	0	0	0	0	0	1	4	4.3
12-1a.m	5	2	5	3	0	0	0	2	3	5	5	1	31	33.3
1-2	0	0	0	1	0	0	0	2	0	0	0	0	3	3.2
2-3	2	3	1	3	0	0	0	1	5	1	2	1	19	20.4
3-4	2	0	0	0	0	0	0	3	0	0	0	1	6	6.5
4-5	1	3	1	0	0	0	0	3	2	1	2	0	13	14.0
5-6	3	0	0	0	0	0	0	0	2	0	1	0	6	6.5
Total	16	15	9	7	0	0	0	11	12	9	10	4	93	100
Relative%	17.2	16.1	9.7	7.5	0.0	0.0	0.0	11.8	12.9	9.7	10.8	4.3	100	

***Anopheles* hourly aggressiveness from 6 pm to 6am. Human Landing Catch Collection Method (HLC). VFT: Vector Feeding Time; 6–11: p.m.; 00–6: am; –: number of mosquito collected.**

Feeding-time of malaria vectors caught by human landing catches at Enugwu-Agidi Njikoka LGA, Anambra State

The feeding time of malaria vectors caught with HLC traps at Enugwu-Agidi showed that 29.6% of malaria vectors fed/rested between 7-8pm followed by 22.7% collected between 12-1am whereas 17.7% of malaria vectors fed/rested between 6-7pm as shown in Table 6.

Table 6: Feeding-time of malaria vectors caught by human landing catches at Enugwu-Agidi

Time	*Numbers of adult mosquitoes caught three consecutive dusk to dawn (Sept. 2021-July. 2022)*													
	Sep	Oct	Nov	Dec	Jan	Feb	Mar	Apr	May	Jun	July	Aug	Total	%
6-7p.m	12	3	4	12	13	3	3	0	0	0	4	0	54	17.7
7-8	9	15	9	17	10	5	9	0	9	0	7	0	90	29.6
8-9	0	0	0	0	0	0	0	0	0	0	0	0	0	0.0
9-10	0	0	0	0	0	0	0	0	0	0	0	0	0	0.0
10-11	0	0	0	0	0	0	2	0	0	0	0	0	2	0.6
11-00	2	2	3	5	1	4	0	0	3	0	2	0	22	7.2
12-1a.m	6	4	10	12	10	12	3	0	6	0	6	0	69	22.7
1-2	2	1	3	0	0	1	0	0	0	0	0	0	7	2.3
2-3	5	1	2	4	2	1	0	0	4	0	0	0	19	6.3
3-4	0	2	1	5	0	0	0	0	3	0	2	0	13	4.3
4-5	3	4	2	2	3	3	2	0	0	0	0	0	19	6.3
5-6	0	0	4	0	2	3	0	0	0	0	0	0	9	3.0
Total	39	32	38	57	41	32	19	0	25	0	21	0	304	100
Relative %	12.8	10.5	12.5	18.8	13.5	10.5	6.3	0.0	8.2	0.0	6.9	0	100	

***Anopheles* hourly aggressiveness from 6 pm to 6am. Human Landing Catch Collection Method (HLC). VFT: Vector Feeding Time; 6–11: p.m.; 00–6: am; –: number of mosquito collected.**

Comparison between seasonal collections of malaria vectors with CDC light traps and HLC Sampling methods

The comparative results of the seasonal collections of malaria vectors using CDC Light Traps and Human Landing Collection (HLC) showed that although malaria vectors were collected during the two seasons of study, nevertheless, a greater number of malaria vectors were collected during the wet season than the dry season in the two collection methods in the three communities of Nibo, Mgbakwu and Enugwu-Agidi. Nevertheless, no malaria vector was caught using the Human Landing Catch method (HLC) in August as shown in Figure 3.

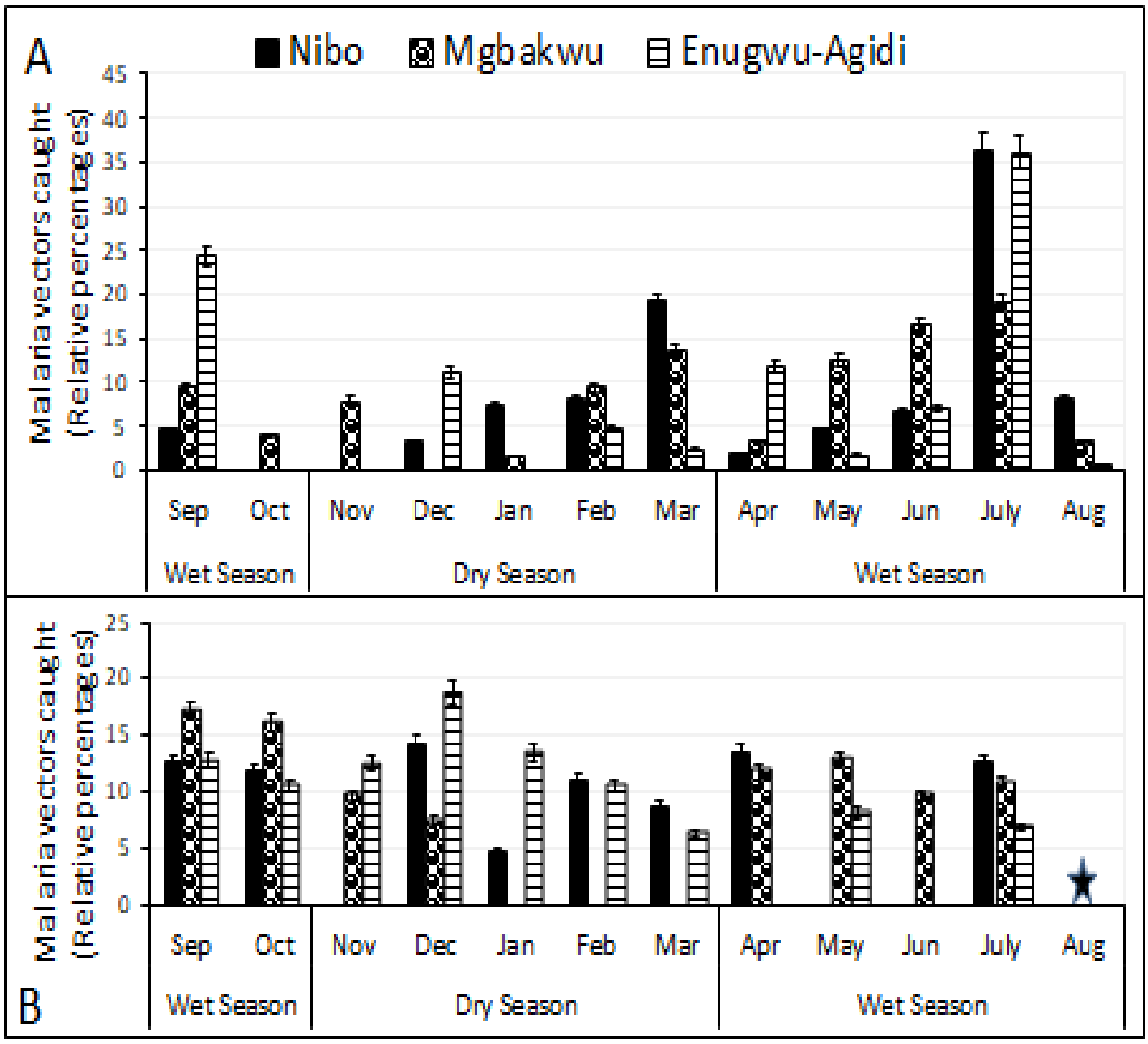


Figure 3: Comparison of seasonal collections of malaria vectors using CDC light traps [A], and human landing catches [B] in three communities from Anambra State, Nigeria

DISCUSSION

It has been observed that insecticide-treated bed nets reduce malaria transmission by limiting contact between mosquito vectors and human hosts when mosquitoes feed during the night (Baton *et al.*, 2005 and Beier, 1998). However, malaria vectors can also feed in the early evening and in the morning when people are not protected as was observed in the feeding and resting patterns of malaria vectors in the three communities of Nibo, Mgbakwu and Enugwu-Agidi from South

East Nigeria. The result was in line with (Eunho *et al*., 2020) who observed in their study that when mosquitoes were maintained under more realistic fluctuating temperatures there was a significant increase in competence for mosquitoes feeding in the evening (6pm) and a significant reduction in competence for those feeding in the morning relative to those feeding at midnight (12 mid-night). These effects appear to be due to thermal sensitivity of malaria parasites during the initial stages of parasite development within the mosquito, and the fact that mosquitoes feeding in the evening experience cooling temperatures during the night, whereas mosquitoes feeding in the morning quickly experience warming temperatures that are inhibitory to parasite establishment (Toure *et al.,* 1994). These results that was referred to indicated the interaction of temperature and feeding behaviour which could be a major ecological determinant of the vectorial capacity of malaria mosquitoes as was also observed in the present study.

The concurrent feeding time of malaria vectors caught with CDC light traps at Nibo, Mgbakwu and Enugwu-Agidi was between 6-7pm and 11pm-12am. This may probably be due to the variations in climatic conditions particularly relative humidity and temperature at each point during sampling. It was also observed that the feeding time of malaria vectors caught with HLC traps at Nibo, Mgbakwu and Enugwu-Agidi was between 2-3am, 12-1am and 4-5am respectively. The result was in lined with the report of the study done by Ngowo *et al.,* (2017) who recorded that variations in household microclimate affect both indoor and outdoor-biting behaviour of malaria vectors in four villages that was sampled where *Anopheles gambiae* feeding peaked at 9-10pm and10-11pm /3-5am at indoor and outdoor points.

Also, Sherrard-Smith, *et al*., 2019 reported in their study that the sample locations had relatively low proportions of malaria vectors feeding when people were indoors and in bed. Also, studies by Saavedra *et al.,* 2019 in riverine communities in the Peruvian Amazon showed that there were more anophelinae collected consistently in the peridomestic area outdoors compared with inside houses. Likewise, Sriwichai, *et al*., 2015 collected more malaria vectors indoors than outdoor when people were likely in beds as was observed in this study. Furthermore, results obtained in this study showed that malaria vectors were caught both indoors and outdoors which may possibly be due to the fact that collection was conducted during both dry season (hot period) and wet season. The comparative result between the two collection methods showed that the relative percentage of malaria vectors caught using CDC Light Trap method where high between 6pm and 12am when compared with the relative percentage of malaria vectors caught between 12am and 6am using Human Landing Catch (HLC) method in the hourly collections. This result showed that malaria vectors do not have a preferred feeding time rather they feed all through the hours from early dusk hour to dawn. Correspondingly, Keven, *et al.,* 2019 in a study on female *Anopheles* in malaria-endemic area of Papua New Guinea showed no significant variation in the abundance of hourly collections using different methods of sample collections.

The indoor feeding and the outdoor feeding behaviour of malaria vectors in the present study was almost the same which makes the occupants living in those houses more quick at risk of infection of the malaria vector. This result was in contrast to the findings of Atting *et al*. (2016) who reported higher feeding behaviour of mosquitoes indoor than outdoor in Uyo, Akwa Ibom State, Nigeria. The endophilic and exophilic behaviour demonstrated by *Anopheles* species in the present study

should be taken into consideration when planning control strategies. Residual house spraying will, therefore, have great impact in areas with a predominant of the mosquito population since the targeted vector will spend enough time on sprayed walls to pick up the lethal insecticide dose. Furthermore, even in the absence of behavioural or physiological resistance, typical biting patterns for many malaria vectors still span periods of the evening and morning when effective coverage of bed nets is less (Killeen, 2014 and Sherrard-Smith *et al*., 2019). This crepuscular biting behaviour contributes to 'residual transmission,' which is the transmission that persists after achieving full universal coverage with an effective intervention such as LLINs, to which local vector populations are fully susceptible (Lambrechts, 2011 and Durnez, 2013). Though insecticide-treated bed nets reduce malaria transmission by limiting contact between mosquito vectors and human hosts when mosquitoes feed during the night, malaria vectors can also feed in the early evening and in the morning when people are not protected and this fact should be taken into considerations when planning for effective and efficient malaria vector interventions.

Conclusion and recommendation

In conclusion, the study showed that CDC light trap and Human Landing Catch collection methods are very effective and has high efficiency in trapping of malaria vectors. Malaria vectors were equally caught both in the outdoors and indoors due to the twelve months longitudinal periods of sample collection which covered both the dry and wet seasons of the year. Malaria vector feeding habit in relation to time also shows that the vectors are non-specific in their feeding time, recording high peak feeding time at 6-11pm and 1-6am in all the study areas. The heterogenic nature and behavior of malaria vectors has caused a lot of hindrances to malaria control strategies and interventions deployed as noticed in this research work. The findings of this study also indicated that the entomological parameters of malaria transmission which includes their resting and feeding behavior, species distribution, presence of sporozoites and other parameters are key pointers in the study areas. This may help narrow down vector control measures in these communities of Awka South, Awka North and Njikoka Local Government Areas of Anambra State to a targeted and selective ecologically sound and cost effective (malaria) vector control strategy suited to the study areas. The findings from this study shows that there is need for the people of the study areas to be properly and effectively educated on environmental health, sanitation, and the use of LLINs during indoors activities so as to reduce human-vectors contact of malaria vectors.

Author contributions

EOO and DNA wrote the first draft of the paper, developed the protocol, oversaw study implementation and monitoring, while ION did the statistical analysis of the work and REI edited the manuscript.

Ethical approval and informed consent

Ethical approval for the research was obtained from the office of the Director, Nnamdi Azikiwe University Human Research Ethics Committee with Reference number: NAU/HREC/2S/02/12/2023/05. The participants were mobilized by a sensitization rally during which the objectives of the study was explained. Participation was voluntary and participants had the liberty to withdraw from the study. We also confirm that all methods used in the study were performed in accordance with the relevant guidelines and regulations as approved by the ethical committee.

Conflict of interest

None declared.

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