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Abundance of mosquito vectors of human diseases at the Awka campus of Nnamdi Azikiwe University, Awka Anambra state Nigeria

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ABSTRACT

Background: Mosquitoes are blood sucking insects dreaded for their biting nuisance and annoyance; they are also incriminated in the transmission of public health diseases among humans and among animals. The study was conducted to determine the abundance of mosquitos and their biting patterns at the Awka Campus of Nnamdi Azikiwe University (NAU), Awka Anambra State between April and June 2021. **Methods:** Eggs of mosquito species were collected using ovitraps and larvae were sampled across five (5). Indoor-biting and resting mosquitoes were collected using pyrethroid knockdown (PKD) method. Outdoor-biting mosquitoes were collected using Human Landing Catch (HLC) method. **Results:** A total of twenty-one (21) mosquitoes comprising three species, Aedes aegypti 3(14.3%), Aedes africanus 13(61.9%) and Aedes albopictus 5(23.8%) were collected using ovitraps. Mosquito larvae collected from breeding sites were a total of sixty-eight (68). The highest number of mosquito larvae were collected from discarded tyres 54(79.4%). Indoors, a total of one hundred and ten (110) adult mosquitoes were collected, and outdoor adult mosquitoes collected were a total of one hundred and forty-eight (148). The abundance of mosquito species collected was significantly different (p<0.05) across the different sampling methods Human Bait> Insecticide Knockdown> Larvae Collection>Ovitrap. The relative abundance of Aedes aegypti 60(40.5%) was significantly higher (p<0.05) than other species. **Conclusion:** In general, outdoor mosquito biting peak was recorded between 6:45pm and 7:45pm. This study is of public health concern because the student and staff population may be exposed to mosquito bites and possible disease transmission.

Introduction

Mosquitoes are of tremendous public health importance because of their ability to host and transmit various disease pathogens and parasites including viruses, protozoa and nematodes [1]. Among dipterans, mosquitos rank first in the spread of infectious diseases such as malaria, yellow fever, filariasis, dengue fever, encephalitis as well as other viral and bacterial diseases [2,3]. Mosquitoes are regarded as public enemies because of the biting nuisance, annoyance, sleeplessness and allergic reactions due to their bites; in addition to these they also transmit diseases that cause morbidities and mortalities. *Anopheles* species, most *Culex* and *Aedes* species are anautogenous mosquitoes and must necessarily take a blood meal to obtain proteins for egg production [4]. When mosquitos' blood

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feed, the parasites they transmit enter the blood stream of their host and by this blood feeding process, they vector many diseases of public health importance [5]. Therefore, since their first association with the transmission of such pathogens to humans and other vertebrates, targeting mosquito to interrupt disease transmission has been the main control strategy against mosquito borne diseases.

Mosquito vectored diseases have high prevalence in the tropics and in 2020 alone, malaria accrued over 241 million cases globally with 95% of malaria cases and 96% of malaria deaths occurring in sub Saharan Africa, with Nigeria alone accounting for about one-third (31.9%) of mortalities due to malaria [6]. Apart from malaria, *Aedes aegypti* and some other species transmit yellow fever and dengue fever viruses, while *Culex quinquefasciatus* is the primary vector for lymphatic filariasis in semi-urban areas [1].

Mosquitoes have a world-wide distribution, occurring throughout the tropical and temperate regions where warm-moist environmental and climatic conditions favour the breeding of mosquito vectors [7]. Tropical areas, including Nigeria, however have the best combination of adequate rainfall, temperature and humidity allowing for the breeding and survival of mosquitoes. The abundance of *Anopheles gambiae sensu lato* in Nigeria is a major factor for the high prevalence of the malaria in the country. In addition, high longevity and fecundity in *A. gambiae s. l* allows for the completion of several gonotropic cycles during which they lay a large number of eggs [8]. Other strategies for survival by mosquitoes include anthropophily (attraction to humans) exophagy (propensity to bite outdoors), endophagy (propensity to bite indoors) and endophily (having a propensity to rest indoors) [9,10]. In addition, ability to withstand drought and choice of breeding sites all play a role in the relative abundance of mosquitoes in different locations and at different seasons and could significantly impact disease epidemiology [11] . This study is therefore aimed at investigating the relative abundance and biting patterns of mosquito vectors of public health importance at Nnamdi Azikiwe University (NAU) Awka Campus, South East, Nigeria.

Materials and Methods

Study area

The study was conducted at the permanent site of Nnamdi Azikiwe University Awka, Anambra State. Awka is the capital city of Anambra State and is located in the lowland rain forest zone of Southern Nigeria. It is located along Onitsha-Enugu Expressway, Awka and has a population of 37,182. It has a relative humidity of 70% reaching 80% during the rainy season and an annual rainfall of about 2000mm. The daily temperature ranges from 26^0C-35^0C during the dry season and from 22.1^0C-30^0C during the wet season. The Institution is a coeducational and higher educational institution which offers courses and programmes leading to officially recognized higher educational degrees such as undergraduate certificate/diploma, bachelor degrees, master degrees and doctorate degrees in several areas of study. It has a total of 14 Faculties and 87 Departments.

Collection of mosquito eggs

Mosquito eggs were collected using ovitrap. The trap consisted of a plastic cup half-filled with water and lined internally with a strip of white calico forming a complete ring within the cup [12]. Five traps each were set at 5 different locations and left for 48 hours before collection. At collection, the strips of the white cloth were carefully removed from the containers, examined for mosquito eggs and air-dried at room temperature for 3-7 days. The eggs on the calico strips were soaked in white plastic bowls covered with mosquito nets and held in place with rubber bands. The hatched out larvae were reared to adult for easy identification [13].

Collection of larval and pupal stages of mosquitoes

Mosquito larvae and pupae were collected from different mosquito breeding sites including discarded tins, motor vehicle tyres and plastic containers around the schools using dippers and a transparent plastic bucket. The collections were done between 8:00am and 11:00am. The larvae and pupae were transferred into specimen bottles with the aid of pipette. The specimen bottles were covered with mosquito nets to prevent the larvae from suffocating. The collected larvae were reared to adult in cages [13].

Collection of indoor-biting adult mosquitoes

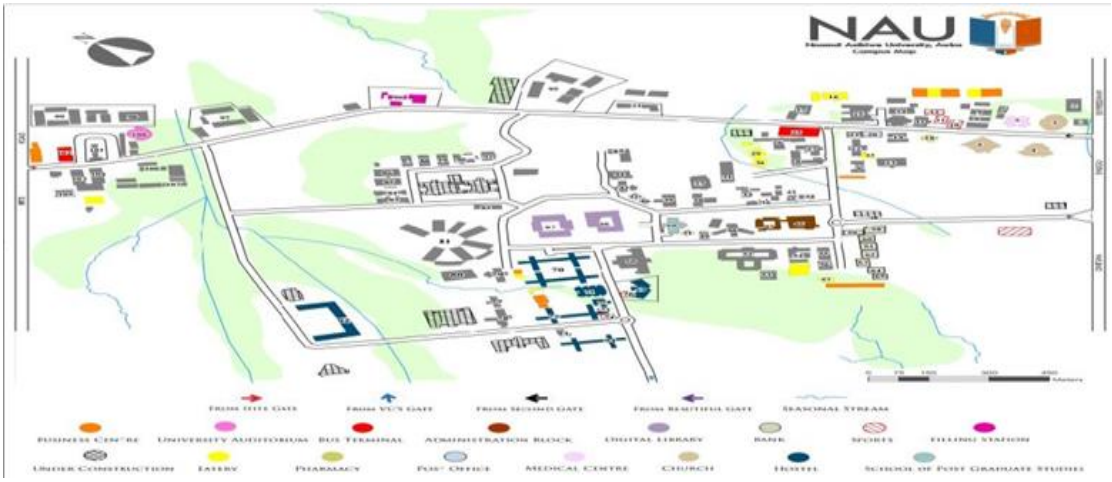
Indoor-biting (endophagic) and resting (endophilic) adult mosquitoes were collected between 6:00am and 8:30am using insecticide (pyrethrum) knockdown collection method [14].

Collection of outdoor-biting adult mosquitoes

Outdoor-biting adult mosquitoes were collected using human landing catch (HLC) method. Four

volunteers served as bait, the collection was done between the hours of 5:00pm and 8:00pm for three 3. Volunteers were made to wear shorts to expose their legs to mosquito's, as much as possible all other parts of their bodies were covered with protective clothing. Volunteers sat on low stools about three meters apart from each other. With the aid of a very bright torchlight, mosquitoes alighting (landing on) the body of the volunteers were stunned by the bright light and collected in test tubes which were immediately stopped with cotton wool and placed in labeled cellophane bags to differentiate time of collection. All catches made were recorded at quarter-hourly intervals (15 minutes each). Mosquitoes caught were transported to the laboratory for identification by species using morphological keys [2]. The laboratory used for the entire research experiment was Parasitology and Entomology Laboratory, Nnamdi Azikiwe University, Awka.

Figure 1. Map of Nnamdi Azikiwe University (NAU) Awka, Anambra State.



Source: Department of Surveying and Geoinformatics, NAU

Data analysis

Chi-square (χ^2) was used to determine the association between breeding sites and mosquito species. One-way Analysis of Variance (ANOVA) was used to determine the differences between the breeding sites, traps and mosquito species. The biting rates of the outdoor-biting mosquitoes, was expressed as: the total number of mosquitoes captured/ no of persons /no of hours/ number of days [15].

Results

A total of three hundred and forty seven (347) mosquitoes were sampled, the mosquitos comprised of four genera (*Aedes, Culex, Anopheles* and *Mansonia*) and seven species. *Aedes aegypti* 117(33.72%) had significantly higher total abundance (p<0.05) than the other species. The abundance of mosquito species collected via the different sampling methods was significantly different (p<0.05) human landing catch> insecticide knockdown> larvae collection>ovitrap. Ovitrap and larvae collection yielded 3 species each while human landing catch (HLC) method yielded twice as many 6 mosquito species as shown in **figure (2)**.

Five breeding sites were visited, namely: ground pools, plastic containers, discarded motor vehicle tyres, blocked gutters/ ditches and tin cans as shown in **figure (3)**. The highest number of mosquito larvae 54(79.4%) was collected from discarded motor vehicle tyres while plastic containers and tin cans yielded equal numbers of larvae 7(10.3%).

The landing pattern of the mosquitoes at quarter-hourly intervals of collection is shown in **figure (6)**. Mosquitoes were collected at every quarter-hourly interval during the HLC experiment 5.00 - 8.00pm West African Time (WAT). The population of mosquitoes landing on the collectors fluctuated within the intervals giving rise to multiple peaks of landing. Total mosquitos collected increased significantly (p<0.05) each hour as the day got darker 5pm<6pm<7pm.

The outdoor biting experiment was carried out from (5pm-8pm). The biting rates of the outdoor-biting mosquitoes of each species was calculated as: the total number of mosquitoes captured/ no of persons /no of hours/ number of days (15) as shown in **table (1)**. For *A. aegypti*, a total of 60 mosquitoes were collected, from 4 persons who

were exposed for 3 hours (5-8pm) for 3 days: 60/4/3/3 =1.67. There was significant difference (p<0.05) in biting rate between species with *A. aegypti* recording highest biting rates.

Figure 2. Relative abundance of mosquito species collected with different methods.

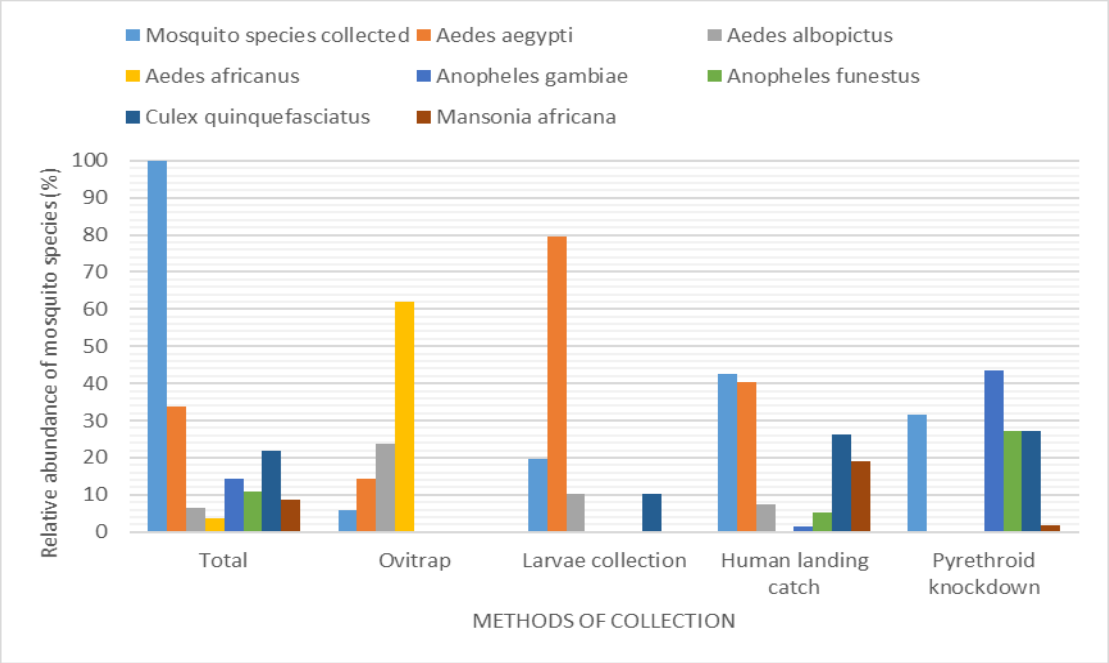


Figure 3. Relative abundance of mosquito larvae collected across different breeding sites.

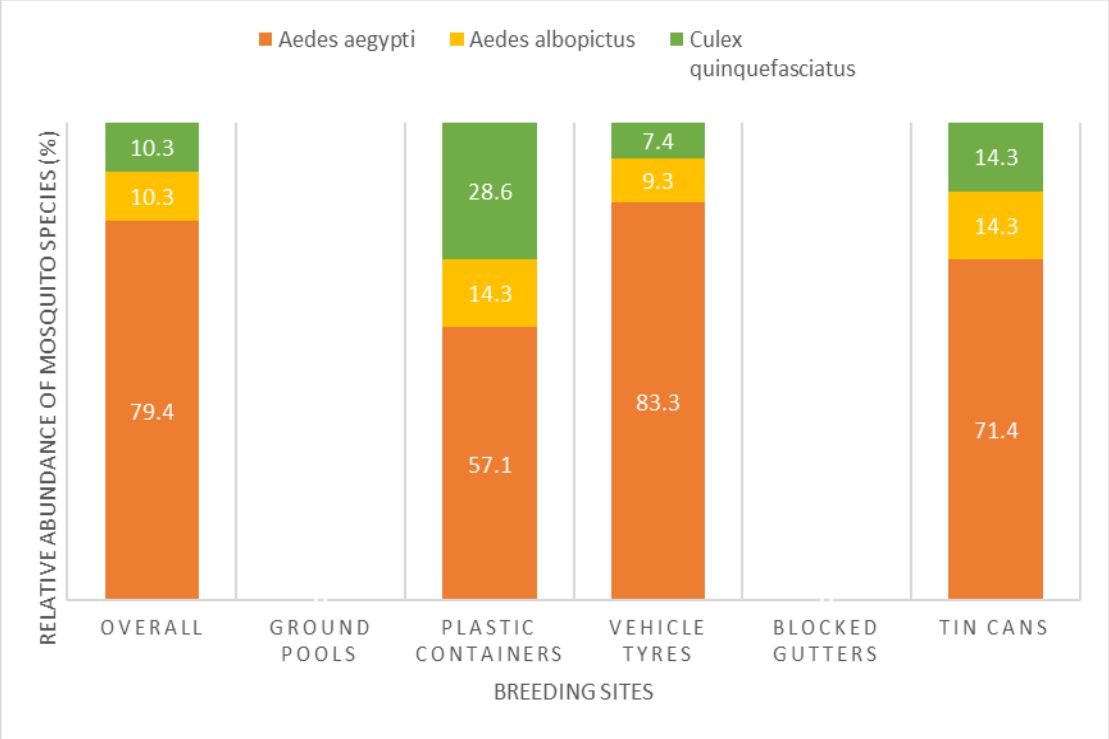


Figure 4. Relative abundance of indoor-biting mosquito species collected with Pyrethrum Knockdown collection method.

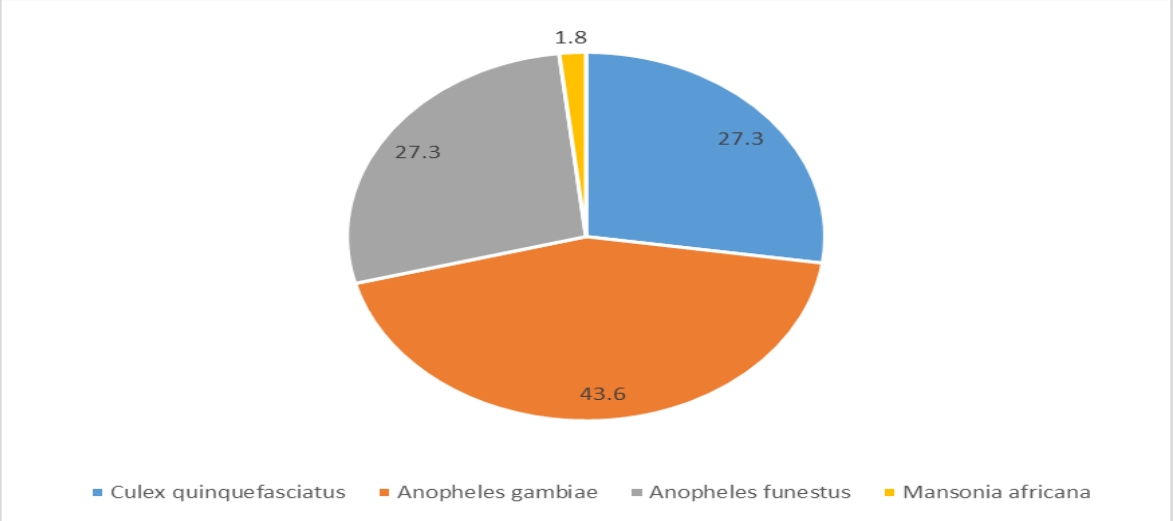


Figure 5. Outdoor-biting mosquito species collected with human bait.

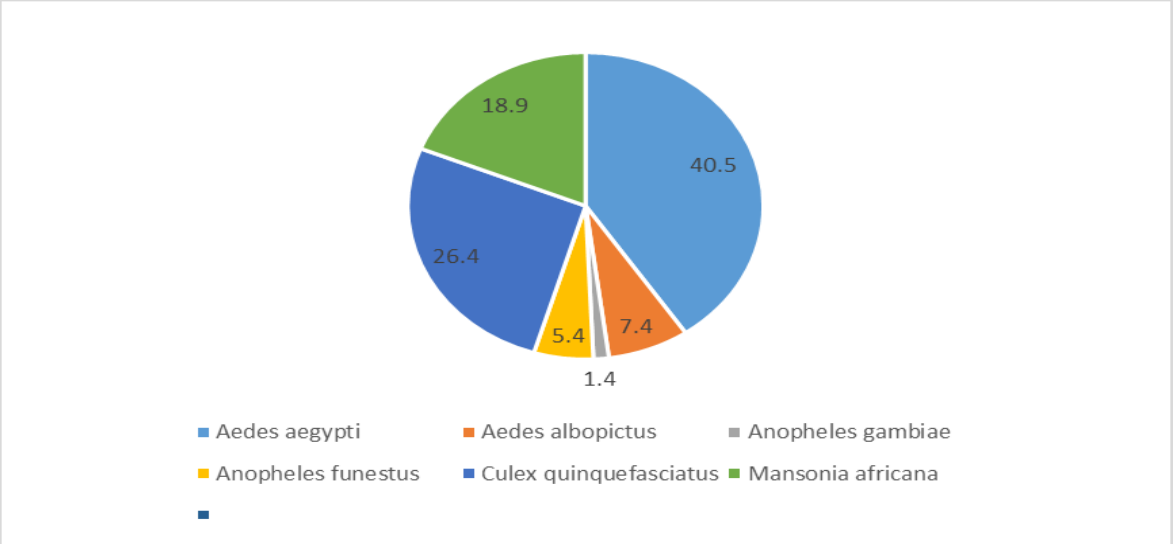


Figure 6. Outdoor-biting mosquito species collected at quarterly hour (15-minutes) intervals .

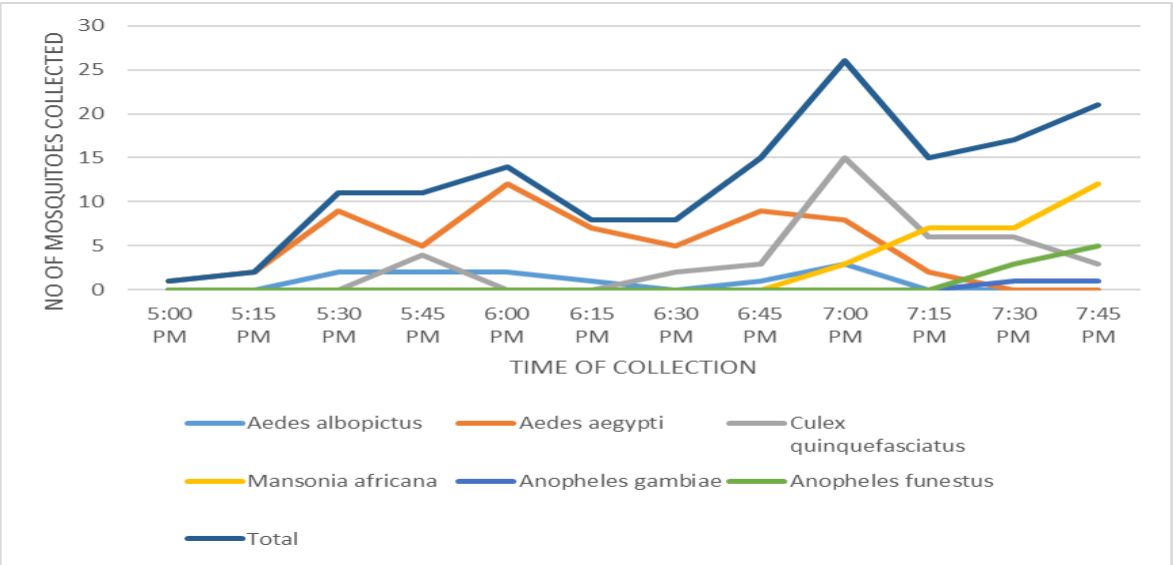


Table 1. Biting rate of different outdoor-biting mosquito species.

Mosquito Species	No Collected	No of Collectors	No of days Collected	Total hours of collection per day	No of man hours used	Biting Rate
Aedes aegypti	60	4	3	12	36	1.67
Aedes albopictus	11	4	3	12	36	0.31
Anopheles gambiae	2	4	3	12	36	0.06
Anopheles funestus	8	4	3	12	36	0.22
Culex quinquefasciatus	39	4	3	12	36	1.08
Mansonia africanus	28	4	3	12	36	0.78

Discussion

A total of 7 mosquito species were collected within NAU University premises in the course of this study with *A. aegypti* having the highest total/overall relative abundance. This could be as a result of the season (end of dry season/early onset of rainy season) in which the research was carried out. *Aedes aegypti* is known to be able to withstand drought and as such they continue to breed even during dry and harsh weather conditions. *Aedes* species where the only species collected using ovitrap, this is consistent with the fact that *Aedes* species are container breeders and can take advantage of small quantities of water to lay their eggs which may hatch to larvae when flooded with water. *Anopheles* mosquitoes in contrast are selective breeders and oviposit exclusively in sunlit, flood pools or swamps.

In a similar research, the presence of 6 species of mosquitoes across 4 genera in Federal University Lafia, North Central, Nigeria towards the end of the rainy season [16]. **Akunne et al.** however reported 4 genera and as many as 13 species in NAU during the rainy season in 2008, which the author attributed to the presence of thick bushes and forest areas as well as building and borehole construction activities ongoing at the time [17]. Our findings of *Aedes* having the highest relative abundance were in agreement with **Ezihe and Chukwuekezie** carried out an extensive eight-year study (2007-2014] monitoring the yellow fever vector (*Aedes* species) in Nigeria, and reported that *A. aegypti* and *A. albopictus* had significantly increased in abundance in in south east Nigeria [18]. This comes as no surprise considering the regular outbreaks of yellow fever being reported in the region.

Mosquito larvae were only found in three of the five larvae sampling sites visited. And only three mosquito species were collected from the three breeding sites that contained mosquito larvae. This could be attributed to the environmental conditions of the habitat in the study area as at the time of the study, rainfall had been irregular and pools dried off before the next rain. Thereby limiting the amount of available breeding sites for mosquitoes. Only larvae of *Culex and Aedes* genera were found across the breeding sites with *Aedes aegypti* being the most abundant species of mosquitoes. The occurrence of more man-made breeding sites (such as containers and discarded tyres) in the campus has accounted for the high prevalence of *Aedes* species.

Aigbodion et al. [19] found both *Aedes* and *Culex* species breeding in artificial containers that are contaminated with organic matters. *Aedes aegypti* is particularly known for its preference for man-made receptacles [12, 20]. The highest number of mosquito larvae was collected from motor vehicle tyres. Reyes-Villanueva *et al* reported that used vehicle tyres contribute significantly to high abundance of *A. albopictus* [21]. *Aedes egypti* is believed to have been imported into Nigeria from Asia and America through 'second-hand' tyres trading [22]. The absence of *Anopheles* caught from the breeding sites could be attributed to the low patterns of rainfalls during the study period which does not allow collection of ground pools which favours the breeding of *Anopheles* mosquitoes.

Outdoor biting peaks occurred as the day got darker. It is therefore advised that people as much as possible avoid resting outdoors at dusk to reduce the chances of being bitten by mosquitoes

and contracting the diseases they vector. The outdoor biting mosquitoes collected were *Aedes, Culex, Anopheles* and *Mansonia*. *Aedes* species mainly bite in the day and at twilight hours when their victims are relaxing after the day's activities. They also indiscriminately bite animals [23] as they are mainly exophagic feeding on both animals and humans and they have been incriminated in the transmission of zoonotic diseases. Culicine mosquitoes were mainly collected outdoors than indoors showing preference for exophagy over endophagy. *Anopheles* on the other hand were more abundant indoors than outdoors. Our findings were in agreement with **Ezihe et al.** who reported the presence of *Anopheles gambiae s. l* and **Umeaneto et al.** who reported the presence of both *Anopheles gambiae s. l and* culicine mosquitoes indoor NAU hostels [24,25]; However neither of the studies scope included a comparism of both indoor and outdoor abundances. **Olayemi et al.** on the other hand argued that while *Anopheles* species are mainly anthropophilic, they are capable of adjusting their feeding habits to when and what host/bloodmeal is available [26].

It is important to stress that all the mosquitoes collected in both larval and adult sampling are efficient vectors of one or more deadly and debilitating tropical diseases. Therefore, the results of the present study have serious public health implications on the humans living inside the campus. In addition to malaria that is ranked as the most deadly tropical disease in Africa, *A. gambiae* has been described as an efficient transmitter of elephantiasis, a debilitating disease caused by a filarial nematode. *Culex quinquefasciatus and Mansonia africanus* are also known to be important in the transmission of elephantiasis in urban areas [27]. In north-central Nigeria, after 8 years of mass drug administration with Albendazole and Ivermectin for the control of lymphatic filariasis, mosquito vector continued to show low-grade infection rates until the distribution of long-lasting insecticidal nets which complimented disease control efforts in the area. This is important as presently Anambra State is in the process of controlling/eliminating lymphatic filariasis, gleaning from Jos and Nasarawa State (North-central) experience, it is advised that additional efforts should be made towards an integrated control approach with emphasis on mosquito vector control to avoid recrudescence.

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Conflict of interest: None.

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