

MALARIA VECTOR ABUNDANCE AND THE INCIDENCE OF MALARIA PARASITE AMONGST STUDENTS LIVING IN NNAMDI AZIKIWE UNIVERSITY HOSTELS

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ABSTRACT

Introduction: Malaria remains an important public health disease in both tropical and subtropical countries of Africa where transmission is majorly through the bite of an infected female *Anopheles* mosquito.

Aim: The main aim of this study was to determine the prevalence of malaria parasite and the abundance of malaria vectors in and around the university hostels.

Place and Duration of study:—The study was carried out in some selected hostels of Nnamdi Azikiwe University, Awka, from the month of March to September, 2018.

Methodology: One hundred and fifty (150) students which volunteered from the selected hostels were tested for malaria parasite. Questionnaire on the frequency of clinical symptoms were distributed to these students. Indoor resting mosquitoes were collected through pyrethrum spray-sheet collection (PSC). Larval sampling was carried out for the immature stages of mosquitoes.

Results: Of the students examined for malaria parasite, 135 (90%) showed positivity to the parasite. From the questionnaire distributed, 30 (20%) of the students treat malaria every 3months, 20(13.3%) every 6 months, 24 (16%) before resuming school, 76(50.7%) only when they develop clinical malaria. Two hundred and two (202) mosquito larvae were collected from their breeding habitats during larval sampling and two hundred and four (204) adult mosquitoes of different species were collected indoors. Although there was no significant difference existing between the number of rooms sampled and the number of mosquitoes collected at 5% level of significance ($\chi^2_{tab} > \chi^2_{cal}$; 9.488 > 6.307).

Conclusion: Re-echoed awareness on what malaria infection is, its prevention and control through personal and environmental management should be given to the students as they go for medicals on school registration.

Keywords: Malaria, Incidence, Students, Vector abundance

INTRODUCTION

Malaria remains an important public health disease in both tropical and subtropical countries of Africa where transmission is principally through the bite of an infected female *Anopheles* mosquito (1). Transmission rarely occurs through direct inoculation of infected red blood cells through blood transfusion, congenital transfer or sharing of needles (2).

Malaria prevalence in an area is mostly determined by the indigenous anopheline mosquitoes, its relative abundance, feeding pattern, resting behaviour and the suitability of human host to *Plasmodium* parasite, among others (3). At least forty-one species of *Anopheles* have been incriminated as vectors of human malaria (4). The most competent of these vectors are found in Africa and belong to the *Anopheles gambiae* and the *Anopheles funestus* species complexes (5).

The National Malaria Control Program (NMCP) in a bid to controlling malaria in Nigeria has delivered about 17 million Insecticide Treated Nets (ITNs) during 2005-2007 (6.6 million Long Lasting Insecticidal Nets), enough to cover only 23% of the population at risk (6). The programme delivered 4.5 million single dose packages of ACT in 2006 and 9 million in 2007, far below total requirements. Funding for malaria control was reported to have increased from US\$17 million in 2005 to US\$60 million in 2007, provided by the government, the Global fund and the World Bank. Though the control of malaria has witnessed a little success but the issue of inadequate malaria epidemiological data (6), drug resistance and vector resistance to insecticides (7, 8), widespread of presumptive treatment (9), incorrect diagnosis (10) and lack of government support on entomological surveillance (11) has increasingly challenged the control and apparently elimination of malaria in Nigeria.

The government though, has been committed to malaria control by intensifying the malaria awareness campaign, emphasizing prevention and eradication of malaria using effective malaria control programme for pregnant women and children of pre-school age (12). This control programme is yet to be extended to higher institutions of learning as malaria is known to have a negative impact on performance of students (13). Based on the considerable number of youths in different universities across the country, government needs to show good commitment in the provision of good health care system especially in the aspect of malaria control programmes in universities. As expected, students absent from lectures for one week or more over a semester due to malaria parasite infection have a higher possibility of poor school performance than those who were absent for less than one week for other genuine reasons. School absenteeism due to malaria parasite may lead to loss of knowledge provided in the lecture hall, leading to students academically lagging behind other students in the same class (14). The use of free or subsidized chemotherapy and insecticide treated nets (ITNs) needs to be extended to the academic communities.

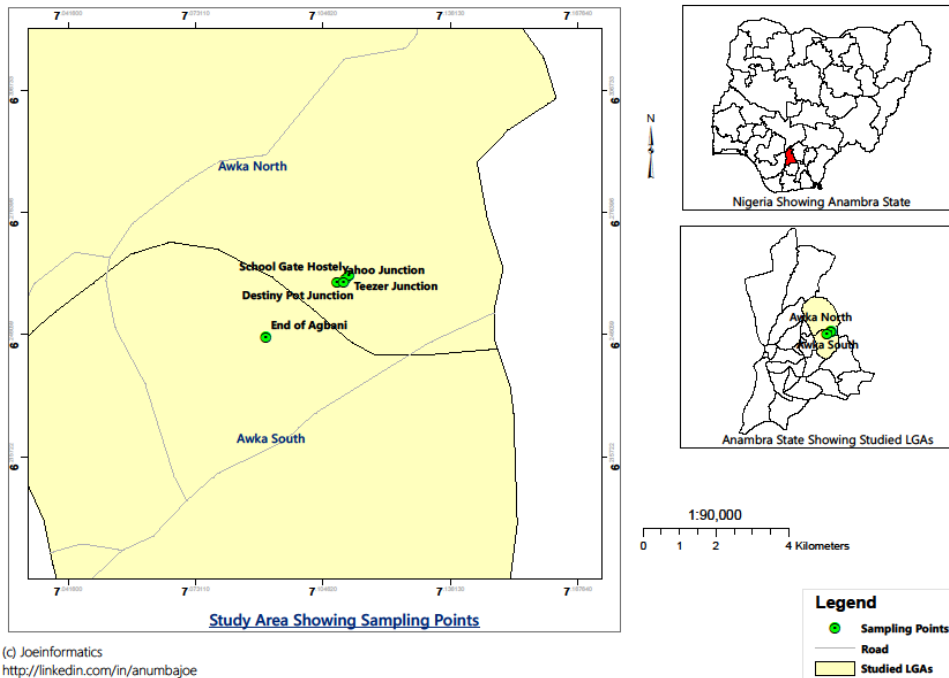
The main aim of the study was to determine the prevalence of malaria parasite and the abundance of malaria vectors within the various locations in and around the university hostels.

Study Area

The study was carried out in the school hostels of Nnamdi Azikiwe University, Awka, Awka-South local government area of Anambra state from March to September, 2018. The climatic condition during the study period created favourable breeding sites for *Anopheles* species which are the known vectors of *Plasmodium* parasites. The study area and its environment houses mainly the students and workers in Nnamdi Azikiwe University, Awka.

TABLE 1: SAMPLE COLLECTION POINTS

LOCATIONS	DESCRIPTION	LANDMARK	GEOGRAPHICAL CORDINATES
Location A	(Hostels before the school)	Commissioner's quarter Ifite	06.2459N, 007.0997E
Location B	Hostels in front of the school)	Yahoo junction	06.2590N, 007.1080E
Location C	(Hostels after the school)	Teezer junction	06.26067N, 007.1109E
Location D	(Hostels farther down the school)	Next level junction	06.2608N, 007.1108E
Location E	(Hostels at the end of Agbani-Ifite).	St Stephen's junction	06.2665N, 007.1202E



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Figure 1: Map of the study site

Study Population

The participants in the study were students of the University living in hostels in Agbani-Ifite. One hundred and fifty (150) students which volunteered from the selected hostels were tested for malaria parasite and the fifty (50) rooms randomly selected were also sampled for indoor resting mosquitoes. All of the participants in the study were healthy individuals who did not show any common signs of malaria.

Ethical Consideration

Ethical approval was obtained from the Dean of the Student Affairs, Head of Department Parasitology and Entomology, Nnamdi Azikiwe University. Verbal consent was obtained from the occupants of all rooms used. Provision were made to contain students who may be absent on the supposed testing date. Anti-malaria drugs were given to students with positive parasitemia as recommended by World Health Organization.

Sample &Data Collection

Samples were collected from five different locations based on the population of the students using the school gate as stand point: Landmarks of the location of the hostels were listed in terms of the junctions closest to the hostels while the geographical coordinates used were for the particular hostels used for the study. Mosquito samples were collected indoors using the pyrethrum spray sheet collection (PSC) technique and larval sampling while blood samples collected from students volunteers that lives in the selected hostels were collected and tested for malaria parasite. Questionnaires were used to collect the bio-data and clinical details of the participants which volunteered. Their bio-data such as names, ages and sex were collected through oral interview and recorded in a field note book. The participants were aged between 17-30 years and were subdivided into 3 age groups of three years intervals each.

Blood samples Collection, film preparation and Examination

Capillary blood samples of the participants were collected carefully into an EDTA container using a 2ml syringe. The samples were used to make thick blood films on clean grease free slides. The smear was allowed to air-dry and then properly stained with Giemsa stain. After air-drying the stained smear it was viewed under a light microscope for presence of *Plasmodium* species. Interest was not on speciation of *Plasmodium* infestation but on the presence and degree of parasite in the blood sample of each participant

Mosquito larval collection

Larvae from the different selected locations were collected and put in labelled sampling containers. Collected samples were differentiated based on certain macroscopic features such as their movements and the presence or absence of siphon and were identified using the Identification keys of (15).

Collection of indoor-biting mosquitoes

Enumerated hostel rooms were sampled for indoor biting and resting adult mosquitoes using the pyrethrum knockdown technique. Edibles and water were taken away from the rooms before the exercise. White sheet of cloth were spread on the floor of the rooms and precautions were taken to ensure that the windows and doors were properly shut for each room at the sampling time. The rooms were sprayed with Insecticide (Raid^{TR}), a brand of domestic aerosol insecticide commonly available in the local markets.

After 20 minutes of spraying each room, the doors and windows were opened and the cloths were folded. Folding of the cloths was from the edges to ensure that all knocked down mosquitoes concentrated at the centre. They were then taken to the open space outside where they were opened. All the mosquitoes were carefully picked with forceps into Eppendorfs tubes.

Identification Of Mosquito Samples

The collected mosquitoes were mounted on glass slides and viewed under microscope for identification using relevant taxonomic keys (5). Morphological features of the mosquitoes such as, palps, proboscis, wings, scutellum, legs, thorax, abdomen, size and colour were used to identify the adult mosquitoes (16).

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Administration Of Questionnaires

Structured questionnaires were given to randomly selected students of the various sampled hostels to provide information on how often they treat malaria, the drugs and insecticide used in the treatment of malaria and what they use for prevention of mosquito bite respectively, if self treatment and confirmatory testing is practiced, and what factors determine their choice of drugs as well as their use of interventions such as insecticide treated nets.

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Data Analysis

The data were analyzed using: tabulations, percentages, and test of statistical significant differences using Chi square (X^2). The statistical software used was the Statistical Program for Social Sciences (SPSS) Version 23. The level of significance was taken at $P = 0.05$.

Results Of Blood Samples Examined

Of the 150 participants whose blood samples were examined, 135 persons representing 90% of all the participants were infested with *Plasmodium* at varying degrees. It was observed that all the students residing at Location E had different degrees of *Plasmodium* infestation, showing 100% prevalence. Though, students residing at Location B revealed a lower *Plasmodium* infestation than those residing in other areas, it was still a very high prevalence rate of 76.7%. The cumulative presence of *Plasmodium* in each location was recorded and the total prevalence rate tabulated. The difference in prevalence of malaria in the various locations was statistically significant at 5% level of probability ($x^2 \text{ cal} > x^2 \text{ tab}$; $16.88 > 9.488$)

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TABLE 2: PREVALENCE OF MALARIA INFECTION IN DIFFERENT LOCATIONS

Locations	Numbers examined	Numbers infected	Prevalence rate (%)
Location A	30	26	86.7
Location B	30	23	76.7
Location C	30	29	96.7
Location D	30	27	90
Location E	30	30	100
Total	150	135	90

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Malaria parasite among the students examined showed that males (68) are more prevalent to the parasite than the females (67). Although there is no statistical significant difference between prevalence of malaria infection among males and females at 5% level of significance ($\chi^2_{tab} > \chi^2_{cal}$; 3.841 > 0.0473). Ages 22-26 were more prevalent to the parasite (94.3%), although no significant difference between the parasitemia and the age groups at 5% level of significance ($\chi^2_{tab} > \chi^2_{cal}$; 3.841 > 0.0473). From the blood samples examined, It was also observed that males within the age bracket of 22-26 years had the highest prevalence rate of 96% (Table 3).

TABLE 3: PREVALENCE RATE OF MALARIA PARASITE ACROSS DIFFERENT AGES AND SEXES

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Age group	Total number examined		Number positive		Prevalence %		Overall Prevalence %
	Female	Male	Female	Male	Female	Male	
17-21	28	24	24	23	85.7	95.8	90.4
22-26	28	25	26	24	92.8	96	94.3
27-30	19	26	18	20	94.7	76.9	84.4
Total	75	75	68	67	90.7	89.3	90

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Result Of Mosquito Collections

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A total of two hundred and two (202) mosquito larvae were collected from different breeding habitats which include: pot holes, stagnant water, gutters, hoof marks and tyre marks. One hundred and forty five (145) *Anopheles* larvae were collected from natural pools of water and eighty eight (88) were of *Culex* species. On emergence to adult, only one hundred and eighty five (185) emerged to adult. Of the emerged adult, one hundred and thirty seven (137) were *An.gambiae* sl and forty eight were *Culex* species.

TABLE 4: LOCATIONS OF LARVAL SAMPLING, EMERGED SPECIES AND HABITAT CONDITIONS

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Types of Breeding site	Locations	Species collected	Number	Characterization of breeding sites	Number of Mosquitoes Emerged
pot holes, gutters, foot marks and tyre marks	A	<i>An.g</i>	13	clear water with algea	17
		<i>Cx.que</i>	4		
pot holes	B	<i>Cx.tig</i>	4	mud water with leaves, gutters	<i>Cx tigripes</i> larvae fed on 9 <i>Cx .que</i> before they were separated 16 <i>Cx que</i> , 4 <i>Cx tig</i> and 2 <i>An.g</i>
		<i>Cx.que</i>	25		
		<i>An.g</i>	2		
pot holes, gutters, tyre marks	C	<i>Cx.que</i>	24	dirty smelly water where run-off water from hostels collect	24
pot holes, gutters, foot marks and tyre marks	D	<i>An.g</i>	42	water with gasoline, fermented water from cassava, drums for collection of rain water	34 larvae emerged as 8 were fed upon by predators
Farms, potholes, gutter	E	<i>An.g</i>	88	dirty water with leaves as shades	88

Note: *An.g* = *Anopheles gambiae* sl, *Cx. que* = *Culex quenequefasciatus*, *Cx. tig*= *Culex tigripis*

A total of 50 rooms were sampled from among the selected hostels and 3 different species of different genera were collected. Although there was no significant difference existing between the number of rooms sampled and the number of mosquitoes collected at 5% level of significance ($\chi^2_{tab} > \chi^2_{cal}$; 9.488 > 6.307). Nevertheless, there was a significant number of *Anopheles* mosquitoes collected from the

different locations. Among the mosquitoes collected, 48 (23.5%) were *Anopheles gambiae* sl, 147 (72.1%) were *Culex quiquefasciatus* and 9(4.4%) were *Mansonia africana*.

TABLE 5: INDOOR RESTING ADULT MOSQUITOES COLLECTED IN THE HOSTEL USING PYRETHRUM SPRAY SHEET COLLECTION (PSC)

Locations	Number of room	Number of mosquitoes	Mean <i>Anopheles</i> abundance	Species of mosquitoes collected		
				<i>An gambiae</i> sl	<i>Cx quiquefasciatus</i>	<i>Mansonia africana</i>
A	13	36	0.3	4(11.1%)	32(88.9%)	0 (0%)
B	13	30	0.8	8(26.7%)	22(73.3%)	0(0%)
C	12	55	1.4	17(31.5%)	37(68.5%)	1(1%)
D	12	35	0.4	7(16.7%)	25(83.3%)	3(1%)
E	12	48	1	12(27.9%)	31(72.1%)	5(%)
Total	50	204	3.9	48(23.5%)	147(72.1%)	9(4.4%)

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Result From Questionnaires

From the questionnaires shared to the 150 students, it was gathered that 30(20%) of the students treat malaria every 3months, 20(13.3%) every 6 months, 24 (16%) before resuming school, 76(50.7%) only when they fall ill to malaria.

On the possession of LLINs, majority {81 (54%)} of the students have LLINs, while the remaining {69(46%)} students do not. Out of the 81 (54%) of the students that have the nets, 34 (54%) of them do not use it, 20(24.7%) use it every day, 8(9.9%) use it only when they remember, 4(4.9%) use it only when they are cold and 15(18.5%) of them use it anytime they like.

DISCUSSION

The results obtained in the study showed that the prevalence of malaria infection among Nnamdi Azikiwe university students living in Agbani-Ifite is high (90%). The result is in line with the findings of (17) conducted among first year students of the same university. The finding in this study is relatively high when compared to 38.93% by (16) among students living in the university hostel, 61% for students in University of Abuja by (18), and 59.4% recorded for post primary students of Umunede and Asaba by (19), though most of the studies were carried out in the wet season.

The high malaria infection prevalence could be attributed to the night activities of the students such as parties, ranging from birthday parties to departmental nights. The most common of these activities is night reading, normally practiced by majority of the students and more often done in the open (Garba square) thereby exposing themselves to mosquito bites. Also in some hostels in Location E, the rooms were overcrowded with an average of 6 to 8 students per room thus making use of mosquito net as preventive measure useless. It is worrisome that many of the students do not have mosquito nets and among those that have, only (24.7%) of them use them judiciously (everyday) attributed to the high prevalence. A few, 4(4.9%), of the study population use the LLINs only during cold weather conditions which is in agreement with the findings of (20).

From our findings, higher prevalence rate were seen in males than females: out of 75 males, 68(90.7%) of them were infected, and of the 75 females sampled, 67(89.3%) of them were infected although there was no significant difference between prevalence of malaria between males and females. This agrees with the finding of (21) who reported that prevalence of malaria among the male gender is higher than of the female. (22) also reported that the cause of higher prevalence observed in male could be due to the fact that they expose their bodies more than females when the weather is hot and thus increases the chances of being bitten by the mosquito. The sampled male students most often exhibit a carefree attitude and pay little or no attention to damaged window nets, bad doors and the use of mosquito nets compared to the female students. Ages 22-26 of the students sampled were more prevalent to the parasite (91%) than other ages as (12) observed the same. The reason might be that many of the students in this age bracket are either in their final years or close to that and they want improve on their performance in school as to come out of school with a better result so they increase in their reading at night while others amongst the age bracket wants to make their last days in school count by hanging out, visit drinking spots and clubs only to come back very late at night.

From the study of (23), it was observed and likewise in this study that malaria transmission is not homogenous but dependent on two primary factors; location of the breeding sites and clustering of human habitations where people are serving as reservoirs of malaria parasites. Locations C, D, E have many breeding sites for *Anopheles* mosquitoes consisting of pools and puddles of various sizes majority of which were created artificially. Of the larvae collected in the breeding sites, 62% were *Anopheles* larvae. More collections were made in Location E with dirty water which has leaves as shades. The collection of *Anopheles gambiae* larvae in dirty water supports the finding of (24) that some species of *Anopheles gambiae* mosquitoes can also breed in dirty water as against the general knowledge that *Anopheles* mosquitoes breeds in a clean and clear water (25). The breeding sites with smelly dirty water collected more *Culex* mosquitoes. Surprisingly, *Anopheles* mosquito larvae were collected in the habitat with gasoline at location D, though it has also been reported in the study of (26). This mechanism of *Anopheles* larvae surviving under oily breeding sites have been observed in the study of (27) that it enhances selection pressure which ends with the emergence of adult population capable of withstanding the lethal doses of pyrethroid which is the mainstay of *Anopheles* mosquito control though we did not study this mechanism. The result of the study showed that *Anopheles gambiae* breeds in almost all the locations where students live, thus every person is at risk of malaria attack. Attributes of rate of exposure

of the study population to vector bites due to their activities in school and the environment forms a factor in the transmission pattern of the disease. Owing to the un-tarred roads and constructions going on in the study area, potholes, foot marks and many tyre marks were created allowing standing water that served as favourable breeding sites for the malaria vector and other mosquito vectors.

Poor drainage system and waste disposal was obvious especially at locations D and C where water from various rooms collects through exposed gutters from the different rooms channelled to the major collecting gutter; and majority of the students here do not use mosquito nets. The use of interventions were highly practiced by the students in locations A and B especially from location B coupled with the fact that sampled hostels from this location were mainly neat hostels with clean surroundings. Hence corresponding to *Anopheles* mosquito abundance in the rooms sampled with malaria prevalence, there was a relationship as seen in Locations D and E. The difference in malaria prevalence and number of *Anopheles* mosquitoes in these locations was significant ($P=0.014$). The study of (28) opined that since the *Anopheles gambiae* species is a very effective vector of malaria, the presence of even one is a big cause for public health concern; thus, the effect of forty-six of them in an area cannot be over emphasized.

Differences in mean *Anopheles* mosquito abundance in the various locations could be due to the pattern of the room and the number of persons living in the rooms. More mosquitoes were collected from rooms occupied by larger number of persons (3-8 students) compared to rooms occupied by a single student as (29) observed. This is because *Anopheles gambiae* sl are strongly attracted to the scent from the human body, as observed by (30) that the more the people in a room the higher the concentration of the scent and a corresponding increase in the number of mosquitoes attracted as was seen in location E.

In conclusion, the study have shown how highly exposed the university students living in the hostels are to malaria infection and a host of other mosquito borne diseases. There is a need for re-echoed awareness to the students as they go for medicals on school registration what malaria infection is, its prevention and control through personal effort and environmental management.

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Competing Interest

There was no conflicting interest

AUTHORS' CONTRIBUTIONS

Conceived and designed the experiments: Ezihe Ebuka Kingsley, Egbuche Chukwudi Micheal, Ukonze Chikaodili. Performed the experiments: Ezihe Ebuka Kingsley, Egbuche Chukwudi Micheal, Ukonze Chikaodili and Nwankwo Edith. Analyzed the data: Anumba Joseph Uche, Umenzekwe Chukwudi Christian. Blood collection and testing for malaria parasitemia: Ogudu Emmanuel Ogbonnia. Contributed reagents/materials/analysis tools: Anumba Joseph Uche. Contributed to the writing of the manuscript: Ezihe Ebuka Kingsley, Egbuche Chukwudi Micheal. All authors read and approved the final manuscript.

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