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## **Malaria Parasitaemia Association with ABO Blood Types among Students of a Private University in Western Delta, Nigeria**

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### **Authors' contributions**

*This work was carried out in collaboration between both authors. Every aspect of this work carried out by author SFL was closely supervised by author FDO in the University laboratory. This also included the write up. Both authors read and approved the final manuscript.*

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### **ABSTRACT**

**Aim:** The focus of this work was to investigate any relationship between ABO blood types and malaria parasitaemia among students of a private University based in Western Delta, Nigeria.

**Study Design:** Whole blood samples were obtained from a randomly sampled number of students and dispensed into ethylene-diamine-tetra-acetic acid (EDTA) containers which were appropriately labelled. Collected blood samples were tested for ABO blood types and malaria parasites by standard methods. Data obtained were statistically analyzed.

**Place and Duration of Study:** The study was carried out in the Microbiology and Biotechnology laboratory of Western Delta University, Oghara, Nigeria between May, 2013 to October, 2013.

**Methods:** Venous blood of 2ml volume was obtained by venepuncture from 360 students made up of 150 (41.7%) males and 210 (58.3%) females of 28years average and who were both symptomatic and asymptomatic for malaria due to *Plasmodium falciparum*. Malaria parasite screening was done by both *P. falciparum* antigen rapid (Micropoint, USA) test and Giemsa staining. ABO blood typing was done using Monoclonal Antisera

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A, B and D. Results obtained were analyzed for any association by chi-square statistical method.

**Results:** One hundred and forty one (41.6%) male and 198 (58.4%) female samples were rhesus positive. Nine (42.9%) and 12 (57.1%) males and females respectively were rhesus negative. ABO blood group frequency occurrence was 55.8 % ( O), 22.5 % ( A), 18.3 % ( B) and 3.4 % (AB). A total of 255 (70.8%) students were infected with *P. falciparum* parasites of which 55.3% and 44.7% were females and males respectively. ABO blood group malaria parasitaemia frequencies were 76.4 % (O), 56.3% (B), 52.4% (A) and 0.0% (AB) for non-severe malaria and 70.5% (O), 58.3% (A), 55.6% (AB) and 50.0% (B) for severe plasmodiasis. Whereas there was significant association between malaria infection and gender ( $P < 0.05$ ), there was no significant association between severe and non-severe malaria parasitaemia in relation to ABO blood types ( $P > 0.05$ ).

**Conclusion:** The presence of rhesus negative factor up to 5.8% suggested a gradual and steady rise in the frequency occurrence of the factor when compared to reports of earlier studies. ABO blood groups O and AB recorded the highest and lowest frequencies respectively. The highest parasitaemia rate was observed among group O individuals and also among female O individuals compared to male O individuals. More female than male students suffered from both severe and non-severe forms of plasmodiasis. There was no significant association of all ABO blood types with severe and non-severe malaria parasitaemia clinically implying that all ABO blood types are equally at risk and therefore, available malaria prophylactic and therapeutic strategies should be directed at subjects of all groups.

*Keywords: Malaria infection rates; association; ABO blood types; students; University.*

## 1. INTRODUCTION

Malaria (which is caused by *Plasmodium* species transmitted by anopheles mosquitoes) is the second leading disease following Acquired Immunodeficiency Syndrome-AIDS [1] associated with high morbidity and mortality through anaemia, cerebral complications and other mechanisms [1].

Despite the high morbidity and mortality, certain individuals living in malaria endemic regions appear relatively protected compared to those who suffer frequent severe malaria attacks. Resistance to malaria infection is dependent on the development of an immune response by the host and to a varying extent, on a certain innate characteristic possessing protective value against infection [2]. These factors include Sick cell trait (HbAS) and Sick cell disease –HbSS [3], ABO blood types [4] and the level of G-6-P- dehydrogenase [5].

It is thought that an understanding of the nature of relationship (if any), between ABO blood groups and malaria parasitaemia should provide an invaluable window in the scourge and that studies of malaria parasitaemia from that stand point in populations of malaria endemic regions will be helpful in elucidating any such relationship [6].

A high percentage of severe malaria cases has been reported among blood group A individuals [7,8]. Furthermore, Migot-Nabias [9] and Pathirana et al. [10] observed low paraitemia and uncomplicated malaria cases among blood group O individuals. Similarly, a significant advantage of a link of *Plasmodium falciparum* with ABO blood groups with respect to group O has been reported by some authors [11,12,13]. Other studies have shown high frequency of malaria episodes among blood group A individuals as compared with other

blood group individuals [14]. Akhigbe et al. [15] reported that blood group AB persons had the lowest malaria attack while blood group A persons had the highest attack.

Studies to investigate any possible association between ABO blood group system and some disease conditions have been carried out by some authors [16,17,18,19,20,21]. Some of these studies reported significant associations thereby suggesting that ABO blood groups have an impact on the infection status of the individuals possessing a particular ABO blood group [18,19,20,21].

The absence of significant association between *P. falciparum* prevalence and ABO antigens has also been reported by some studies [1,22,23,12]. Other authors are less equivocal in relating malaria parasitaemia to ABO blood groups [24,25]. Otajevwo [26] in a study associating malaria parasitaemia with ABO blood types among residents of Warri, Nigeria, reported that 6.9%, 19.0%, 20.7% and 53.3% of a total of 174 whole blood samples processed belonged to blood types AB, B, A and O respectively and 138(79.3%) of total sample size were infected with malaria parasites of which *P. falciparum* was the predominant species. In the report, the highest malaria parasite load was observed among group O (52.2%) individuals while the least was noticed among blood group AB (8.7%) individuals. Malaria parasitaemia was higher among the males (83.3%) than females (75.0%). Otajevwo [27] also reported ABO blood types among students of Igbinedion University, Okada, Nigeria of which out of 104 samples analysed, 44(3.9%), 16(15.4%), 32(30.8%) and 52(50.0%) students occurred in AB, A, B and O groups respectively. On the whole, 80(76.8%) of total samples processed, were positive for malaria parasitaemia.

Nkuo-Akenji [28] reported that blood group O individuals maybe more susceptible to malaria attack. A study conducted in Edo State University, Nigeria reported that blood groups O and B male individuals were the most and least susceptible to malaria attack respectively [4]. In a study conducted on inhabitants of Odakpu, Anambra State, Ilozumba and Uzozie [6] reported ABO blood group prevalence of 2.63%, 12.05%, 21.1% and 63.8% for groups AB, B, A and O respectively. Zerihun et al. [29] reported that A, B and AB blood group individuals are more vulnerable to *P. falciparum* infection compared to blood group O individuals in a study they carried among 269 febrile Ethiopian out patients who visited a health care.

Despite the above researches, a consensus on possible association between ABO blood group genes and malaria infection is still lacking [30]. This might be due to limited data or unreported data on the cell antigens [28], peculiar demographic distributions and characteristics of study areas [22]. A definite statement on the apparent trend can be made therefore, if results of more studies on ABO blood groups associated with malaria infection in different parts of Nigeria and other neighbouring countries are made available and accessible [6].

This work is a response to the call for further research in the area and it is aimed at studying ABO blood type association with malaria parasitaemia among students of a private University based in Western Delta, Nigeria with the following objectives:

1. Determine the sex distribution of malaria parasitaemia among students recruited for the study.
2. Determine the distribution of malaria parasitaemia among ABO blood group types of male students.
3. Determine the distribution of malaria parasitaemia among ABO blood types of female students.

4. Determine the distribution of severe malaria parasitaemia among ABO blood types of male students.
5. Determine the distribution of severe malaria parasitaemia among ABO blood types of female students.

## 2. MATERIALS AND METHODS

### 2.1 Ethical Clearance

Informed consent was given by students recruited for the study. A consent form (in form of a questionnaire) was given to students who were all adults to fill and sign. Participating students also gave their verbal informed consent by presenting themselves for blood sample collection.

### 2.2 Sampling

Two millilitres (2ml) of venous blood was collected from 360 randomly selected Western Delta University students made of 150(41.8%) males and 210(58.2%) females from various departments of the University. Blood samples were collected with the aid of 5ml sterile needles and syringes by venipuncture (by tying a tourniquet around the upper arm and sterilizing it with 70% ethanol to increase blood pressure in the veins) and dispensed into ethylene diamine tetra acetic acid (EDTA) anticoagulant bottle containers. Blood and blood containers were properly mixed by standard method and labelled appropriately. Both symptomatic and asymptomatic subjects were used for the study. Symptomatic subjects were those that were positive for malaria parasite test and showed obvious signs of illness. Asymptomatic subjects were those who were apparently healthy although positive for malaria parasite test. For clarity, symptomatic subjects were grouped as those having severe malaria parasitaemia while asymptomatic subjects were those with non-severe malaria parasitaemia.

### 2.3 Malaria Parasite Detection Tests

The Malaria *Plasmodium falciparum* rapid test Device (whole blood ) Micropoint package insert kit by Cook et al. [31] was used to test for the presence of specific antigens (proteins) produced by *falciparum* malaria parasites [32,33]. Antigen targeted is *P. falciparum* specific histidine rich protein-2 (i.e. Pf HRP-2) and aldolase [34,32].

### 2.4 Rapid (Micropoint, USA) Test Procedure

The test device, specimens, buffer and /or controls were allowed to equilibrate to 15-30°C (room temperature) prior to testing. The test device cassette was removed from the foil pouch and used immediately. Using the sample loop (or rubber pipette) provided, 5ul (0.05ml) of whole blood was dropped into sample well (A). Two drops (60ul or 0.06ml) of assay buffer were put in well B and the set up was left to stand for 20minutes after which result was read. A positive result was one in which a pink or purple coloured control (C) band appeared in addition to a distinct pink coloured band in the test (P.f) region. In a negative result, only one pink or purple band appeared on the control (C) region while no apparent band appeared on the test (P.f) region. The quality control region was to confirm if sufficient specimen volume was used and if the correct procedural technique was applied.

## 2.5 Sensitivity of Test Device

The Malaria P.f Rapid (Micropoint) test device (whole blood) has been tested in comparison with the thick blood smears of clinical samples. The results show that the sensitivity of the test device (whole blood) is >99.0% relative to blood smears.

## 2.6 Specificity

The test device uses an antibody that is highly specific for Malaria *Plasmodium falciparum* antigen in the blood. The test device has a specificity of >99.0% relative to blood smears

## 2.7 Expected

The test device has been compared with traditional thick or thin blood smears microscopic analysis. The correlation between the two systems is >99.0%.

## 2.8 Limitation

The test device is for in-vitro diagnostic test use only. Neither the qualitative value nor the rate of increase in Pf antigen concentration can be determined by its qualitative test. The test Device will only indicate the presence of *Plasmodium falciparum* antigen and hence, the specimen should not be used as the sole criteria for the diagnosis of malaria infection. All results must be interpreted together with other clinical information available to the physician. A negative result does not at any time preclude the possibility of malaria infection.

## 2.9 Giemsa Staining

This manual staining was carried out as quality control of the rapid test method. Thick blood films were made on grease free microscope slides (after appropriate labelling and allowed to air dry on laboratory working bench. Slides were arranged on a staining rack and flooded with 10% (v/v) Giemsa stain solution for 15minutes [35]. The plus system was used for the determination of parasite density [36]. Only positive results with the rapid test device and Giemsa staining method were recorded and used for ABO group typing.

## 2.10 Typing Blood Samples for Blood Group Antigens

The ABO blood group of each subject was determined using cell grouping Antisera according to methods described by Rosenfield [37] and Cheesbrough [38]. Monoclonal Antisera A, B and D (Agape Diagnostics, Ltd, India) were used.

## 2.11 Statistical Analysis of Data

Chi square ( $\chi^2$ ) analysis using test of independence of two characters or associations using a 4 x 3 contingency table at 95% confidence interval was used. The software used was the statistical package for social sciences (SPSS) version 17.0. Confidence interval at 95% (0.05) was calculated using mean  $\pm 0.05 S_x$  where 6 refers to degree of freedom calculated using (r-1) (c-1).

### 3. RESULTS

In Table 1, the sex, department and age group distribution of students recruited for the study are shown. Students of fifteen (15) departments gave their informed consent and volunteered their blood samples as shown. Seventy two (20.0%), 66(18.3%), 54(15.0%), 33(9.2%), 30(8.3%), 30(8.3%), 18(5.0%), 18(5.0%), 12(3.3%), 6(1.7%), 6(1.7%), 6(1.7%), 3(0.8%), 3(0.8%) and 3(0.8%) volunteering students of Microbiology/Biotechnology, Accounting, Geology, Economics, Biochemistry, Political Science, Mass Communication, Business Administration, Computer Science, Chemistry, Physics, Library science, Philosophy, Hotel/Tourism and Environmental Science departments respectively engaged of in this study are also shown in that descending order. Out of the 360 subjects sampled, 150(41.7%) and 210(58.3%) were males and females respectively. Table 1 also shows the age brackets of students used for the study. In decreasing order, 198(55.0%), 111(30.8%), 39(10.8%), 6(1.7%), 3(0.8%) and 3(0.8%) students occurred in the 21-25(average age 23years), 15-20(average age 19years), 26-30(average age 28 years), 31-35(average age 33years), 36-40(average age 40years) and 41-45 (average age 44years) age brackets. Again, out of the total recruited in each age group, 90(45.5%), 33(29.7%), 15(38.5%), 6(100.0%), 3(100.0%) and 3(100.0%) represented male students in 21-25, 15-20, 26-30, 31-35, 36-40 and 41-45 age brackets respectively in that descending order while 108(54.5%), 78(70.3%), 24(61.5%), 0(0.0%), 0(0.0%) and 0(0.0%) represented female students in the same age groups respectively.

Table 2 shows data on ABO blood groups and Rhesus factor frequency distribution among students studied. A total of 360 students' blood samples made up of 150(41.5%) and 210 (58.3%) males and females respectively were processed for ABO blood typing. Out of this sample size, 141(41.6%) male samples were rhesus positive. Nine (42.9%) and 12(57.1%) male and female samples respectively were rhesus negative. In terms of male ABO blood grouping, 6(4.0%), 18(12.0%), 36(24.0%) and 90(60.0%) male students were grouped into blood groups AB, B, A and O respectively in that ascending order. With regards to the female students, 6(2.9%), 45(21.4%), 48(22.9%) and 111(52.9%) female students were grouped into blood groups AB, A, B and O respectively in that ascending order.

Table 3 shows the sex (gender) distribution of malaria parasitaemia among students engaged in the study. Out of the 360 students recruited, 150(41.7%) and 210(58.3%) were males and females respectively. Out of the same sample size, 255(70.8%) students were infected (positive) with *P. falciparum* parasites in their peripheral blood circulation of which 114(44.7%) and 141(55.3%) were infected male and female students respectively. Table 3 also shows that 105 (29.2%) students were negative for malaria parasites in their blood system of which 36(34.3%) and 69(65.7%) were uninfected male and female students respectively. Malaria parasitaemia was significantly associated with female gender and therefore with sex ( $\chi^2$  p-value =3.841, cal. Value= 3.958,  $p < 0.05$ ).

**Table 1. Sex and Age Distribution OF Western Delta University Recruited For the Study**

Department	Sex	15-20 Average=19 n=111(30.8%)	21-25 Average=23yrs n=198(55.0%)	26-30 Average=28 n=39(10.8%)	31-35 Average=33 n=06(1.7%)	36-40 Average=40 n=03(0.8%)	41-45 Average=44 n=03(0.8%)
Geology	M=36	06	27	00	03	00	00
n=54(15.0%)	F=18	12	03	03	00	00	00
Microbiology	M=21	12	09	00	00	00	00
n=72(20.0%)	F=51	09	33	09	00	00	00
Accounting	M=24	09	12	03	00	00	00
n=66(18.3%)	F=42	15	24	03	00	00	00
Mass comm.	M=03	00	00	00	00	03	00
n=18(5.0%)	F=15	00	06	00	00	00	00
Computer. Sci	M=06	00	03	03	00	00	00
n=12(3.3%)	F=06	03	03	00	00	00	00
Bus. Admin	M=06	00	03	03	00	00	00
n=18(5.0%)	F=12	00	12	00	00	00	00
Biochemistry	M=03	00	03	00	00	00	00
n =30(8.3%)	F=27	21	06	00	00	00	00
Economics	M=18	03	15	00	00	00	00
n=33(9.2%)	F=15	03	09	03	00	00	00
Pol. Science	M=21	03	12	03	00	00	03
n=30(8.3%)	F=09	06	03	00	00	00	00
Env. Science	M=03	00	03	00	00	00	00
n=03(0.8%)	F=00	00	00	00	00	00	00
Chemistry	M=03	00	03	00	00	00	00
n=06(1.7%)	F=03	00	03	00	00	00	00
Physics	M=03	00	00	03	00	00	00
n=06(1.7%)	F=03	00	00	03	00	00	00
Hotel & tourism	M=00	00	00	00	00	00	00
n=03(0.8%)	F=03	00	03	00	00	00	00
Philosophy	M=03	00	00	00	03	00	00
n=03(0.8%)	F=00	00	00	00	00	00	00
Library sci.	M=00	00	00	00	00	00	00
n=06(1.7%)	F=06	00	03	03	00	00	00
	M=150(41.7%)	33(29.7%)	90(45.5%)	15(38.5%)	06(100.0%)	03(100.0%)	03(100.0%)
	F=210(58.3%)	78(70.3%)	108(54.5%)	24(61.5%)	0(0.0%)	0(0.0%)	0(0.0%)
<b>TOTAL</b>	<b>360</b>	<b>111(30.8%)</b>	<b>198(55.0%)</b>	<b>39(10.8%)</b>	<b>06(1.7%)</b>	<b>03(0.8%)</b>	<b>03(0.8%)</b>

**Table 2. ABO blood groups and rhesus frequency distribution among students recruited for study**

ABO blood types	Sex	Rhesus Positive (%)	Rhesus Negative (%)	Total
A	M	33(23.4%)	03(33.3)	36(24.0)
	F	39(19.7%)	06(66.7)	45(21.4)
B	M	18(12.8)	00(0.0)	18(12.0)
	F	45(22.7)	03(33.3)	48(22.9)
AB	M	06(4.3)	00(0.0)	06(4.0)
	F	06(3.0)	00(0.0)	06(2.9)
O	M	84(59.6)	06(66.7)	90(60.0)
	F	108(54.6)	03(33.3)	111(52.9)
Total	M	141(41.6)	09(42.9)	150(41.7)
	F	198(58.3)	12(57.1)	210(58.3)
		<b>339(94.2)</b>	<b>21(5.8)</b>	<b>360(100.0)</b>

**Table 3. Sex distribution of malaria parasitaemia among students recruited for Study**

Sex	Positive Malaria parasite (%)	Negative malaria parasite (%)	Total
Males	114(44.7)	36(34.3)	150(41.7)
Female	141(55.3)	69(65.7)	210(58.3)
<b>Total</b>	<b>255(70.8)</b>	<b>105(29.2)</b>	<b>360(100.0)</b>

$\chi^2$  df (1)<sub>0.05</sub>=3.841(P-value), Calculated  $\chi^2$  =3.958, P<0.05

The distribution of malaria parasitaemia and severe malaria parasitaemia among ABO blood types of students' data are shown in Tables 4 and 5. Out of a total sample size of 360, 255(70.8%) students were infected (Table 3) of which 206 (67.5%) had non-severe form of malaria parasitaemia and 49(63.6%) students recorded severe form of malaria parasitaemia. Blood group O students recorded the highest non-severe malaria infection rate (76.4%) as well as the highest severe malaria infection rate (70.5%). Although blood type B students recorded the next highest non-severe malaria infection rate (56.3%), blood type A students recorded the next highest severe malaria infection rate (58.3%) after group O. None of the AB blood type students recorded non-severe malaria infection but 5(55.6%) AB blood group subjects had severe malaria infection. The lowest severe malaria infection rate (50.0%) was recorded for blood group B subjects. Also, whereas 99(32.5%) students cutting across all blood types were uninfected with non-severe form of *P. falciparum* malaria parasites, 28(36.4%) students were uninfected with the severe form. Statistically, there was no significant association of blood types with non-severe form of *P.f* malaria infection ( $\chi^2$  df (3)<sub>0.05</sub>=7.815, cal.  $\chi^2$  =3.408, P >0.05) and there was also no significant association of the blood types with severe form of *P. f* malaria infection ( $\chi^2$  df (3)<sub>0.05</sub> = 7.815, cal.  $\chi^2$  =5.652, P>0.05).

**Table 4. Non-severe malaria parasitaemia distribution among ABO blood types of students distribution**

Malaria status	A	B	AB	O	Total
Infected (%)	33(52.4)	27(56.3)	0(0.0)	146(76.4)	206(67.5)
Uninfected (%)	30(47.6)	21(43.7)	3(100.0)	45(23.6)	99(32.5)
<b>Total</b>	<b>63(20.7)</b>	<b>48(15.7)</b>	<b>3(1.0)</b>	<b>191(62.6)</b>	<b>305(100.0)</b>

$\chi^2$  df (3) <sub>0.05</sub> =7.815(P-value), Calculated  $\chi^2$  =3.408, P > 0.05



**Table 5. Severe malaria parasitaemia distribution among ABO blood types of students distribution**

<b>Malaria status</b>	<b>A</b>	<b>B</b>	<b>AB</b>	<b>O</b>	<b>Total</b>
Infected (%)	7(58.3)	6(50.0)	5(55.6)	31(70.5)	49(63.6)
Uninfected (%)	5(41.7)	6(50.0)	4(44.4)	13(29.5)	28(36.4)
<b>Total</b>	<b>12(15.6)</b>	<b>12(15.6)</b>	<b>9(11.7)</b>	<b>44(57.1)</b>	<b>77(100.0)</b>

$\chi^2$  df (3)<sub>0.05</sub> = 7.815 (p-value), Calculated  $\chi^2$  = 5.652,  $P > 0.05$

#### 4. DISCUSSION

A total of 360 students from fifteen departments of the University were engaged in the study. Of this sample size, 198 (55.0%) were of the 21-25 age category and 23yrs average age. This somewhat suggests that the average age of majority of students in the institution under study is 23 years. This is followed by an average age of 19 years as represented by 111 (30.8%) students who occurred in the 15-20 age bracket. This finding implies that the subjects recruited for this study were mature enough to make informed decision in terms of giving their informed consent to be enlisted for the research.

Findings in this study showed that 339(94.2%) and 21(5.8%) students were rhesus positive and rhesus negative respectively of which 141(41.6%) and 9(42.9%) male students were rhesus positive and rhesus negative respectively. That there were more rhesus positive and rhesus negative females than males may have been a factor of chance. This finding is however in consonance with the report of a previous study [27]. It is to be noted that the 5.8% prevalence of rhesus negative factor appears small in view of the total sample size. This should however not be undermined in view of the far reaching medical implication in terms of child birth and still birth arising from haemolytic disease of newborn (HDN). It also suggests that rhesus negative factor is occurring in increasing frequencies from population to population and hence the need for relevant health care providers as well as Ministry of health to track down people with this factor through compulsory blood typing test Results in this work also reveal that the frequency occurrence of the ABO blood types were 55.8%, 22.5%, 18.3% and 3.4% for groups O, A, B and AB respectively (Table 2). This therefore, confirms that blood group O is the most prevalent while blood AB is the least prevalent and this agrees with the reports of previous studies [39,40,41,42,43,19,44,45,15,29,27].

These findings are not consistent with an earlier study by Agbonlahor et al. [4] which reported groups O and A as the highest and least occurring respectively. These differences may be due to ethnic, racial and geographical differences of various populations studied.

The findings in this work further revealed that 255 (70.8%) of the 360 students sampled were infected with *Plasmodium falciparum* parasites of various densities. This rather high parasitaemia rate suggests that Oghara town maybe hyper endemic for malaria. This parasitaemia rate appears to be low compared to 93.4% obtained in Odoakpu, Onitsha South by Ilozumba and Uzozie [6] and 76.8% obtained in Okada, Edo state by Otajevwo [27] as well as 79.3% and 77.4% parasitaemia rates obtained in Warri by Otajevwo [26] and in Owerri by Mbanugo and Emenalo [46] respectively. Parasitaemia rate in this study appears high however, when compared to prevalence rates of 66.9%, 58.3%, 43.2%, 10.0% and 6.0% obtained respectively for subjects in Ogbomosho by Akhigbe et al. [15], children in Awka by Mbanugo and Ejins [47], coastal dwellers in Lagos by Nebe et al. [48], blood donors in Ibadan by Edington et al. [49] and blood donors in Maiduguri by Ahmed et al. [50].

These results may suggest the existence of regional differences in malaria parasitaemia rates in Nigeria with the western, mid western and eastern areas (represented by Ogbomoso, Warri, Onitsha and Owerri) ranking highest in prevalence rating and the northern area (represented by Maiduguri) occupying the lowest position while Lagos and Ibadan take a middle position. Similar studies should however be undertaken by other authors in other parts of the country before a more definite statement on the apparent trends could be made.

Statistical analysis showed that at 95% C.I, the female students were significantly ( $\chi^2 = 3.841$  p-value, Cal.  $\chi^2 = 3.958$ ,  $P < 0.05$ ) more infected than the males ( $P < 0.05$ ) Table 3. The reason why the females are more prone to malaria attack could not be fathomed immediately but it may be due to the peculiar mode of dressing of the typical African girl/woman that exposes parts of their bodies such as hands and legs to mosquito bites most of the time compared to the males. This finding was in agreement with the report of some earlier authors [15] but inconsistent with reports of some previous studies [4,6,46]. According to Portilo and Sullivan [51], genetic factors could play a role by endowing females with immuno-regulatory potentials to cope better with some diseases.

ABO blood type frequencies of non-severe infection were 76.4%, 56.3%, 52.4% and 0.0% for blood groups O, B, A and AB respectively as obtained in this study. Clearly, blood group O students were the highest infected with non-severe malaria and the least were group A subjects. No AB subjects were infected. High prevalence of malaria infection among group O women has been reported [1]. Finding in this study is not in tandem with the report of Akhigbe et al. [15] which stated non severe malaria frequencies of 85.1%, 46.1%, 40.0% and 33.3% for blood groups O, A, B and AB respectively. Findings in this work show (among others) that group O subjects were the most infected and this is supported by the reports of some earlier authors [7,2,52,53,15,10,9]. Present finding however, disagrees with the reports of Barragan et al. [54], Carlson and Wahlgreen [55], Rowe et al. [56], Udomsangpetch et al. [57] which stated that blood group O individuals usually have low malaria attack because parasitized red blood cells have stronger tendency to form rosettes with uninfected erythrocytes of the A, B, AB blood groups than with those of blood group O.

In this study, malaria infection was divided into severe and non-severe malaria. Infected students whose thick films showed the presence of *P. falciparum* asexual forms such as schizonts, merozoites and who showed obvious signs of malaria infection were grouped as severe. It is well known that pathogenesis of severe malaria is due to the presence of schizonts, merozoites and trophozoites of *Plasmodium falciparum* [58]. The frequency occurrence of severe malaria therefore among the blood types were 70.5%, 58.3%, 55.6% and 50.0% for groups O, A, AB and B respectively (Table 5). Again, group O subjects were the most infected with severe malaria while group B students were the least infected. On the whole, group O subjects were most prone to non-severe and severe forms of malaria. This is not consistent with the finding of Akhigbe et al. [15] which stated that blood group O persons are associated with the highest prevalence of non-severe malaria and a lower prevalence of severe malaria. Whereas group AB persons recorded no single non-severe malaria case, 55.6% of AB individuals were infected with the severe form of malaria. The finding by some authors stating that blood group O seems to confer a certain degree of protection against severe cases of malaria disease is therefore, not in consonance with present finding in that regard [7,8,9,10]. Also, according to Athreya and Coriell [59], malaria is a disease for which an association with ABO blood group distribution seems to exist and which may have played an important role in shaping the prevalence in favour of group O.

Present study also reported that whereas blood type AB students were infected with 55.6% cases of severe malaria, no group AB person was infected with non-severe malaria. This is in agreement with report of an earlier study [15] and inconsistent with that of Ilozumba and Uzozie [6].

The pathophysiological plausibility of an interaction between malaria parasite and red blood cells as well as the potential role erythrocyte surface antigens may play in cytoadhesion of infected erythrocytes and parasite invasion may give credence to a possible association between ABO blood types and malaria infection [2]. Numerous studies have been carried out to determine any possible association between ABO blood groups and malaria [1] and most findings from such studies have been contradictory [1]. Herrera et al. [22] reported that there was no association between ABO blood types and malaria parasitaemia and that the controversial association of these variables previously found in other populations may be due to their peculiar demographic distributions and characteristics.

In this study, attempt was made to associate non-severe malaria and severe malaria parasitaemia with ABO blood types (Tables 4 and 5). Chi-square ( $\chi^2$ ) p value (book value) at degree of freedom (3) and 95% confidence interval was 7.815 as against calculated  $\chi^2 = 3.408$  for non-severe malaria association with ABO blood groups (Table 4). Hence  $P < 0.05$  and therefore, no significant association or relationship was established. As for the severe malaria association with the ABO blood groups,  $\chi^2_{0.05}(3) = 7.815$  (p value) and Cal.  $\chi^2 = 5.652$  and hence,  $P > 0.05$  (Table 5). This implied that there was also no statistically significant association existing between the variables. This lack of significant association in both cases is in disagreement with reports of previous studies which recorded significant associations [29] and consistent with reports of some authors [28,60,61,62,1].

## **5. CONCLUSION**

This study showed that there are more rhesus positive female students than male students in the institution under study. The presence of rhesus negative factor up to 5.8% suggested a gradual and steady rise in the frequency of occurrence of the factor when compared to reports of earlier studies. This has far reaching medical implication in terms of erythroblastosis fetalis (or haemolytic disease of newborn). ABO blood groups O and AB recorded the highest and lowest frequencies respectively. Hence, this finding, lends further credence to earlier related studies that in any given population, the highest number of subjects belong to group O while the least number belong to group AB. A striking revelation that followed was that across all ABO blood types screened for, there were more females than males. This may not have been due to chance as it is similar to reports of earlier studies.

The highest parasitaemia rate was observed among group O individuals and also among female group O individuals compared to group O male students. More female than male students suffered from both severe and non-severe forms of plasmodiasis. There was a significant statistical relationship between sex (gender) and malaria parasitaemia. Also, blood group O subjects had the highest prevalence of non-severe and severe malaria infection and whereas no single AB student recorded non-severe malaria infection, up to 50.0% AB students had severe form of malaria attack. It was not clear which of blood groups A and or B recorded the least of severe and non-severe forms of malaria. Statistically, there was however, no significant association of all ABO blood types with severe and non-severe malaria parasitaemia. This ostensibly implies that all ABO blood groups are equally at risk

and therefore, available malaria prophylactic and therapeutic strategies by health agencies should be directed at individuals of all groups.

## INFORMED CONSENT

All authors declare that both oral and written informed consent was obtained from students (subjects) recruited for this work.

## COMPETING INTERESTS

Authors have declared that no competing interests exist.

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