PREVALENCE OF MALARIAPARASITAEMIA AND ITS ASSOCIATION WITH ABO BLOOD GROUPING AMONG STUDENTS OF IGBINEDION UNIVERSITY OKADA, NIGERIA.

ABSTRACT

A total of 104 asymptomatic malaria parasite carrier students of Igbinedion University, Okada, Nigeria were used for the study. Two milliliters (2ml) of venous blood was collected by venipuncture using 5ml hypodermic needles and syringes. Blood samples were immediately dispensed into Ethylene Diamine. Tetra-Acetic acid (EDTA) anticoagulated containers and mixed appropriately. ABO blood typing using monoclonal Antisera A, B and D was carried out on samples. The malaria Plasmodium falciparum rapid Test Device (whole blood) package insert kit was used to test for presence of malaria parasite in specimens. The 104 samples analyzed were made up of 24(23.1%) rhesus positive males, 76(73.0%) rhesus positive females and 4(3.9%) rhesus negative females. In increasing order, 4(3.9%), 16(15.4%), 32(30.8%) and 52(50.0%) students occurred in blood groups AB, A, B and O respectively. Forty (38.4%) of total group O subjects were infected with various densities of malaria trophozoites. Out of group B individuals, 24(23.1%) were infected. All sampled 4(3.9%) AB
students were infected. On the whole, 80(76.8%) of total samples processed, were positive for malaria parasitaemia. Twelve (11.5%) and 68(65.4%) of total male and female subjects were infected. Malaria parasitaemia seemed to be relatively high across all blood groups with groups O and AB subjects apparently recording the highest and least infection rates respectively. There was statistical significant association between malaria parasitaemia and ABO blood groups of both male and female students (P < 0.05) and between malaria parasitaemia and ABO blood groups of female students only (P < 0.05). The association of malaria parasitaemia and ABO blood groups of male students was not significant (P > 0.05). Despite these results, it is still difficult to make a clear conclusion on the association of ABO blood groups and malaria parasitaemia because a lot of study designs have been conducted invivo.
Introduction

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 certain innate characteristic possessing protective value against infection.

These factors include sickle cell trait (HBAS) and sickle cell disease (HbSS) Alouch (1997), ABO blood type (Agbonlahor et al., 1993; Udomsangpetch et al, 1993) and the level of G-6-P-Dihydrogenase activity (Ruwende and Hill, 1998). The high frequency of haemoglobin S in malaria endemic areas has been attributed to the selection advantage it offers to carriers of the trait against malaria (Derek, 1996).

Among all infectious diseases, malaria has been said to cause the greatest harm to humans (Burnet and white, 1972). Inspite of concerted efforts being made by several countries and agencies to eradicate malaria parasite, malaria remains enigmatic and continues to rank among the foremost killer diseases of our time (Ilozumba and Uzozie, 2009). The disease affects about 500 million people and kills about 2 million, mostly children each year (WHO, 2000). Malaria is endemic in about 102 countries with more than half of the world’s population at risk (Smyth, 1994).

Natives of malaria endemic regions which include countries of West Africa, manifest partial immunity to malaria which has been described as premonition or
concomitant immunity (Brooks et al., 2004). In hyper-endemic areas, even infants do not suffer as much as is sometimes thought from malaria infection because they are passively protected by the mother’s antibodies until they begin to develop some of their own (Chandler and Read, 1961).

Some investigators have expressed the opinion that genotype is a factor in susceptibility to plasmodium species infection in humans (Hill et al., 1992; Ademowo et al., 1995; Omotade et al., 1999) but there is lack of consensus on possible association between ABO blood group genes and malaria parasitaemia (Hill et al., 1992) Ademowo et al., 1995; Fisher and Boone, 1998)

The ABO blood groups consist of A, B and H carbohydrate antigens which can regulate protein activities during infection and antibodies against these antigens (Chung et al., 205; Greenwell, 1997). A number of studies were conducted to investigate the association between ABO blood group system and some disease conditions (Tursen et al., 2005; Kassim and Ejezie, 1982; Opera, 2007; Abdulazeeez et al., 2008; Ndambaa et al., 1997; Blackwell et al., 2002). Some of these studies reported significant associations, suggesting that ABO blood groups have and impact on infection status of the individuals possessing a particular ABO blood group (Opera, 2007; Abdulazeeez et al., 2008; Ndambaa et al., 1997; Blackwell et al., 2002).

In view of a heavy burden placed on human health due to malaria, a good many investigations have been conducted to find out whether or not ABO blood groups antigens are associated with susceptibility, resistance or severity of P.
falciparum malaria. Nonetheless, these studies have reported contradictory results. Some studies reported the absence of significant association between P. falciparum (prevalence, parasitaemia or antibody titre) and ABO antigens (Kassim and Ejezie, 1982; Martin et al., 1979 Bayoumi et al., 1986; Akinboye and Ogunrinade, 1987; Thakur and Verma, 1992; Montoya et al., 1994; Uneke et al., 2006). On the other hand, other studies have shown that high frequency of malaria episodes has been observed among blood group A individuals as compared with other blood groups individuals (Beiguelman et al., 2003).

Large numbers of sever malaria cases were also reported among blood group A individuals (Fischer and Boone, 1998; Lell et al., 1999). Further more; Migot-Nabias (2000) and Pathirana et al. (2005) observed low parasitaemia and uncomplicated malaria cases among blood group O individuals respectively.

According to Nkuo – Akenji et al. (2004), it is possible that the ABO blood type, hemoglobin genotype and the presence or absence of Glu–6-P0₄ dehydrogenase activity may have a cumulative effect on the outcome of malaria parasitaemia and parasite density. Variations in reports in the association of ABO bloods groups and diseases progression of P. falciparum malaria show the complexity of the interaction between the parasite and host immune responses.

The death toll from malaria is highest in children and plasmodia parasites invade erythrocytes, the very cell that expresses the ABO antigens on their surface (Adenigka, 2011). Malaria is thus a disease for which an association with ABO blood group distribution seems most plausible and it may have played an
important role in shaping the prevalence in favour of group (Athreya and Coriell, 1967).

In addition, studies have shown the impact of other red blood cells polymorphisms including haemoglobin (HB) abnormalities such as HbS, HbC, thalassemia and deficiency in erythrocytes complement receptor (CR) or glucose – 6- phosphate dehydrogenase on *P falciparum* malaria susceptibility and severity (Migot- Nabias *et al*., 2000; Pathirana *et al*., 2005; Carlson *et al*., 1994; Rowe *et al*., 1997; Weatherall *et al*., 2002; Cock burn *et al*., 2004). This makes it difficult to make a clear analysis of the association of the ABO blood group and *P falciparum* because so far, most studies have been done invivo.

It is thought that an understanding of the nature of relationship (if any) between ABO blood groups and malaria parasitaemia would provide an invaluable window in the effort to contain the malaria scourge and that studies of malaria parasitemia from that stand point in populations of malaria endemic regions will be helpful in elucidating any such relationship (Ilozumba and Uzozie, 2009). Wood (1975) reported that anopheles mosquitoes tend to bite groups O and B persons in preference to group A persons. It has also been reported that group B confers a selective advantage in relation to malaria infection and partly explains why the distribution of group B tends to correspond with a high degree of malaria endemicity (Alhreya, 1967). Postulations that certain antigens on erythrocyte surfaces which enable classification of blood group into ABO systems are involved in the susceptibility of red cell of species of plasmodium have been
widely documented (miller et al., 1997). Besides, susceptibility to malaria parasitaemia could be ABO blood group dependent as suggested by some earlier investigators (Hill et al., 1992; Ademowo et al., 1995). On the other hands, other investigators were unable to establish significant association between ABO blood group and malaria parasitaemia (Akingboye and Ogunrinade, 1987; Thakur and Verma, 1992).

More recent reports are however, less equivocal in relating malaria parasitaemia to ABO blood group (Senga et al., 2007; Rowe et al., 2008). Otajevwo (1997) in a study associating malaria parasitaemia with ABO blood groups 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 groups 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%).

Nkwo – Akenji et al. (2004) determined the effects of ABO /Rh blood groups on malaria. They reported that the highest malaria rate of 74.5% was seen in group O individuals while the lowest (58.6%) was obtained for group B residents. Mean parasite density (Log_{10} - 1/µl blood) in the various blood groups was not significantly different. They concluded that blood group O individuals may be more susceptible to malaria attack. A study conducted of Edo State
University in Nigeria reported that in males, individuals with blood group O and B were the most and the least susceptible to malaria parasitaemia respectively (Agbonlahor et al., 1993).

Prevalence of malaria parasitemia and its possible association with ABO blood groups was investigated among inhabitants of Odakpu of Anambra State, Nigeria by Ilozumba and Uzozie (2009). They reported ABO blood group prevalence of 2.63%, 12.05%, 21.05% and 63.83% for groups AB, B, A and 63.83% respectively. According to their report, malaria parasitemia prevalence varied significantly (P<0.05) with blood group being highest (100.0%) in blood group AB, 94.85% in group O, 90.63% in group A and 89.47% in group B. They concluded that ABO blood group could be a factor that influences susceptibility to infection by human Plasmodium species.

Otajevwo (1997) screened one hundred and seventy four (174) whole blood samples from Warri based residents for ABO blood groups as they relate to malaria infection. Results obtained showed that 12 (6.9%), 33 (19.0%), 36 (20.7%) and 93 (53.3%) of total samples analyzed belonged to blood groups AB, B, A and O respectively and one hundred and thirty eight (79.3%) of total samples were infected with malaria parasites with groups O individuals having the highest infection rate (52.2%) and group AB the least.

In a study carried out by Nkuo-Akenji et al. (2004) to investigate the effects of ABO/Rh blood groups on malaria among subjects based in Buea, south west province of Cameroon, they reported that the highest malaria prevalence (75.5%)
was recorded for group O individual and the lowest incidence (58.6%) for group B individuals. The concluded that group O individuals may be more susceptible to malaria attack.

The distribution of ABO blood groups and their relationship with *Plasmodium falciparum* malaria among febrile 269 outpatients who sought medical attention at a health center in Southern Ethiopia was investigated by Zerihun *et al.* (2011). They reported that A, B and AB blood group individuals are more susceptible to *P. falciparum* infection compared to blood group O individuals. In a similar study carried out on 489 Zimbabwean patients, blood group A individuals were found to have lower haemoglobin levels and at more risk of coma than patients of other blood groups with the conclusion that severe malaria is associated with blood groups (Fischer and Boone, 1998).

Some authors maintain that malaria is a disease for which an association with ABO blood group distribution seems most plausible and may have played an important role in shaping the prevalence in favour of a particular group (Adenigka *et al.*, 2011). Some strains of *Plasmodium falciparum* preferentially trigger rosette formation depending on the red blood cell group with A and B group cells being more likely to form rosettes (Carlson and Wahlgren, 1992; Udomsangpetch *et al.*, 1993). Fischer and Boone (1998) on the contrary reported that malaria occurs in patients of any blood group and no particular blood group precludes the possibility of severe malaria. There is however, still lack of consensus on possible association between ABO blood group genes and malaria parasitaemia (Hill *et al.*, 1992;
Ademowo et al., 1995; Omotade et al., 1999). This might be due to limited data on the association between malaria and red blood cell ABO antigens (Nkuo-Akenji et al., 2004). Hence, results of more studies on malaria parasitaemia in different parts of the country would be needed before a more definite statement on the apparent trend could be made (Ilozumba and Uzozie, 2009).

It is to further extend the frontiers of any possible association between malaria infection and ABO blood groups that this study aimed at investigating malaria parasitaemia association with ABO blood groups among students of Igbinledion University, Okada was carried out with the following objectives:

(a) Determine the ABO blood groups and Rhesus factor frequency distribution among students of Igbinledion University, Okada, Nigeria

(b) Determine if there is a significant association of the ABO blood groups with malaria parasitaemia

(c) Determine the sex distribution as related to ABO blood groups association with malaria parasitaemia

(d) Determine if there is a significant association of ABO blood groups of male and female students separately with malaria parasitaemia
Materials and Methods

Sampling

Using sterile needle and syringes, two milliliters (2ml) of venous blood was collected from 104 randomly selected Igbinedion University, Okada students made of 24(23.1%) males and 80(76.8%) females. Blood samples which were collected by venipuncture (by tying a tourniquet around the upper arm and sterilizing arm with 70% ethanol to sterilize and increase blood pressure in the veins) were dispensed into ethylene- diamine-tetra-acetic acid (EDTA) anticoagulated blood containers, properly mixed by standard method and labeled appropriately. Subjects used were asymptomatic malaria parasite carriers. All symptomatic subjects were excluded from study.

Typing Blood Samples for ABO Blood Groups

The ABO blood group of each subject was determined using cell grouping antisera according to methods described by Rosenfield (1976) and Cheesbrough (2000). Monoclonal Antisera A, B and D (Agappe Diagnostics Ltd, India) were used.

Malaria Parasite Detection Test

The Malaria Plasmodium falciparum rapid Test Device (whole blood) package insert kit by Maconell (2001) and Cookeetal, (1999) was used to test for presence of malaria parasite in the specimens.
Test Kit Content / Storage

The content of test kit included: test devices, test tubes, package insert, disposable specimen droppers transfer droppers and buffers. The test kit was stored at room temperature on laboratory bench according to manufacturers’ instruction. Test Kit was used within expiration date stated on the sealed pouch.

Rapid Test Procedure

The test device, specimens, buffer and/or controls were allowed to equilibrate to 15-30°C (room temperature) prior to testing for processing of each sample, the test device 9cassette0 was removed from the foil pouch and used immediately. Using the transfer dropper (provided), and held vertically one dropper of whole blood sample approximately 20µl (0.02m1) was transferred to the test tube. Added to this, were three full drops of buffer (approximately 120µl or 0.12m1) after which the mixture was allowed to stand for 1 minute. The test device (cassette) was placed on a clean and level surface. The test tube was squeezed five times to mix the specimen and buffer completely. The entire content (of about 140 µl) was then transferred to the specimen well in the cassette with the transfer dropper and the set up was left for 15-20 minutes.

Interpretation of Results

A test was positive if two distinct coloured lines appeared. One line in the control region (c) and another line should be in the test region (T). The intensity of
the colour in the test line region (T) may vary depending on the concentration of *Plasmodium falciparum* present in the specimen. A test was termed negative if one coloured line only appeared in the control region (C) with no apparent coloured line appearing in the test region (T).

**Quality Control**

Internal procedural controls were included in the test. A coloured line appearing in the control region (C) was used as an internal positive procedural control. The quality control was to confirm if sufficient specimen volume was used and if the correct procedural technique was applied.

**Sensitivity of Test Device**

The Malaria P.F Rapid Test Device (whole blood) has been tested within or thick blood smears on clinical samples. The results show that the sensitivity of the test Device (whole blood) is >99% (greater than 99%) relative to blood smears.

**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 >9% relative to blood smears.

**Expected values**

The Test Device has been compared with traditional thick or thin blood smears microscopic analysis. The correlation between the two systems is >99%.
Limitation

The Test Device is for in-vitro diagnostic test use only. Neither the quantitative value nor the rate of increase in PF antigen concentration can be determined by his 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 any time preclude the possibility of malaria infection.

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 labeling) 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 15 minutes (Brooks *et al.*, 2004).

Statistical Analysis of Data

Chi square ($X^2$) analysis using test of independence of two characters or associations using a 4x3 contingency table at 95% confidence interval was used.
Results

Presented in Table 1 are data representing ABO blood group and Rhesus factor frequency distribution among students of Igbinedion University, Nigeria. A total of 104 students’ blood samples made up of 24(23.1%) males and 80(76.8%) females were processed for ABO blood grouping. Out of this sample size, 24(23.1%) male samples were rhesus positive while 76(73.0%) female samples were rhesus positive. Only 4(3.9%) female samples were rhesus negative. In terms of male ABO blood grouping, 4(3.9%), 8(7.7%) and 12(11.5%) male students were grouped into blood groups B, A and O respectively. There was no male AB group. With regards to the female students, 4(3.9%), 8(7.7%), 28(26.9%) and 40(38.4%) students were grouped into AB, A, B and O respectively (Table 1)
Table 1: ABO blood groups and Rhesus factor frequency distribution among
students of Igbinedion University, Okada, Nigeria.

<table>
<thead>
<tr>
<th>ABO blood Groups</th>
<th>Sex</th>
<th>Rhesus Positive</th>
<th>Rhesus Negative</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>M</td>
<td>8(7.7%)</td>
<td>0.0%</td>
<td>8(7.7%)</td>
</tr>
<tr>
<td></td>
<td>F</td>
<td>8(7.7%)</td>
<td>0.0%</td>
<td>8(7.7%)</td>
</tr>
<tr>
<td>B</td>
<td>M</td>
<td>4(3.9%)</td>
<td>0.0%</td>
<td>4(3.9%)</td>
</tr>
<tr>
<td></td>
<td>F</td>
<td>24(23.1%)</td>
<td>4(3.9%)</td>
<td>28(26.9%)</td>
</tr>
<tr>
<td>AB</td>
<td>M</td>
<td>0.0%</td>
<td>0.0%</td>
<td>0.0%</td>
</tr>
<tr>
<td></td>
<td>F</td>
<td>4(3.9%)</td>
<td>0.0%</td>
<td>4(3.9%)</td>
</tr>
<tr>
<td>O</td>
<td>M</td>
<td>12(11.5%)</td>
<td>0.0%</td>
<td>12(11.5%)</td>
</tr>
<tr>
<td></td>
<td>F</td>
<td>40(38.4%)</td>
<td>0.0%</td>
<td>40(38.4%)</td>
</tr>
<tr>
<td><strong>TOTAL</strong></td>
<td>M</td>
<td>24(23.1%)</td>
<td>0.0%</td>
<td>24(23.1%)</td>
</tr>
<tr>
<td></td>
<td>F</td>
<td>76(73.0%)</td>
<td>4(3.9%)</td>
<td>80(76.8%)</td>
</tr>
</tbody>
</table>
Data on the association of ABO blood groups with malaria parasitaemia are presented in Table 2. A total of 4(3.9%), 16(15.4%), 32(30.8%) and 52(50.0%) students sampled belonged to blood groups AB, A, B and O respectively in that increasing order. From the highest to lowest, 40(38.4%) of the total blood group O individuals were infected with malaria trophozoites (parasites) of which 16(15.4%) students scored a density of (+) to (+++) while 24(23.1%) had scanty trophozoite parasites in their peripheral blood. The next group was blood group B of which 24(23.1%) students were infected. The least infected group was AB group with only 4(3.9%) infected individuals. Incidentally, all the 4(3.9%) AB students sampled were infected. On the whole out of 104(100.0%) sampled whole blood specimens, 80(76.8%) representing more than 50% were positive for malaria parasitaemia of which 28(26.9%) individuals scored malaria parasite densities of (+) to (+++) and 52(50.0%) had scanty parasites in their blood stream.
Table 2: Association of ABO blood groups of Igbinedion University students, Okada with malaria parasitaemia

<table>
<thead>
<tr>
<th>ABO blood Groups</th>
<th>Malaria Parasite Density Test</th>
<th>Total Malaria Positive</th>
<th>Negative</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>+++</td>
<td>+</td>
<td>Scanty</td>
<td></td>
</tr>
<tr>
<td>A</td>
<td>0.0%</td>
<td>4(3.9%)</td>
<td>8(7.7%)</td>
<td>12(11.5%)</td>
</tr>
<tr>
<td>B</td>
<td>0.0%</td>
<td>4(3.9%)</td>
<td>20(19.2%)</td>
<td>24(23.1%)</td>
</tr>
<tr>
<td>AB</td>
<td>0.0%</td>
<td>4(3.9%)</td>
<td>0.0%</td>
<td>4(3.9%)</td>
</tr>
<tr>
<td>O</td>
<td>4(3.9%)</td>
<td>4(3.9%)</td>
<td>8(7.7%)</td>
<td>24(23.1%)</td>
</tr>
<tr>
<td>TOTAL</td>
<td>4(3.9%)</td>
<td>4(3.9%)</td>
<td>20(19.2%)</td>
<td>52(50.0%)</td>
</tr>
</tbody>
</table>

Chi – square ($X^2_{0.05,6}$) = 12.592 ( P or Book value)

Calculated or Critical Value = 13.663

P < 0.05
In Table 3, results on sex distribution as they relate to ABO blood groups association with malaria parasite infection are presented. Out of the 104 (100.0%) sampled population, there were 24(23.1%) males and 80(76.8%) females of which 12(11.5%) of total sampled males representing 50%, were infected while 68(65.4%) out of total sampled females representing more than 50%, were infected. Of the total male subjects, 0.0%, 4(3.9%), 8(7.7%) and 12(11.5%) students were grouped into AB, B, A and O blood groups respectively. There was no male AB blood group subject. There were more female students sampled of which 4(3.9%), 8(7.7%), 28(26.9%) and 40(38.4%) female students were placed into AB, A, B and O blood groups respectively in that ascending order. In group A male individuals, 4(3.9%) out of 8(7.7%) sampled were infected with malaria parasites. All 4(3.9%) group B male subjects sampled were infected with malaria parasites and there were no sampled male AB individuals. Out of 12(11.5%) sampled male blood group O individuals, 4(3.9%) were infected with malaria trophozoites. In blood group A, all the 8(7.7%) female blood samples were infected with malaria parasites and this was similar to the trend in the male subjects of the same group. Twenty (19.2%) out of 28(26.9%) sampled female blood group B students were infected with the parasite and like the case of blood group A, all the 4(3.9%) blood group AB female students were infected with the parasite. Lastly, in blood group O, 36(34.5%) female students were infected out of 40(38.4%) total sampled for the group (Table 3).
Table 3: Sex Distribution as Related to ABO Blood Group Association With Malaria Parasitaemia

<table>
<thead>
<tr>
<th>ABO Blood Groups</th>
<th>Sex</th>
<th>Malaria Parasite Density Test</th>
<th>Total Malaria Parasite positive (i.e. +++ , ++ , + &amp; Scanty)</th>
<th>Negative</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>+++</td>
<td>++</td>
<td>+</td>
<td>Scanty</td>
</tr>
<tr>
<td>A</td>
<td>M</td>
<td>0.0%</td>
<td>0.0%</td>
<td>0.0%</td>
<td>4(3.9%)</td>
</tr>
<tr>
<td></td>
<td>F</td>
<td>0.0%</td>
<td>0.0%</td>
<td>4(3.9%)</td>
<td>4(3.9%)</td>
</tr>
<tr>
<td>B</td>
<td>M</td>
<td>0.0%</td>
<td>0.0%</td>
<td>0.0%</td>
<td>4(3.9%)</td>
</tr>
<tr>
<td></td>
<td>F</td>
<td>0.0%</td>
<td>0.0%</td>
<td>4(3.9%)</td>
<td>16(15.4%)</td>
</tr>
<tr>
<td>AB</td>
<td>M</td>
<td>0.0%</td>
<td>0.0%</td>
<td>0.0%</td>
<td>0.0%</td>
</tr>
<tr>
<td></td>
<td>F</td>
<td>0.0%</td>
<td>0.0%</td>
<td>4(3.9%)</td>
<td>0.0%</td>
</tr>
<tr>
<td>O</td>
<td>M</td>
<td>0.0%</td>
<td>0.0%</td>
<td>4(3.9%)</td>
<td>0.0%</td>
</tr>
<tr>
<td></td>
<td>F</td>
<td>4(3.9%)</td>
<td>4(3.9%)</td>
<td>4(3.9%)</td>
<td>24(23.1%)</td>
</tr>
<tr>
<td>TOTAL</td>
<td>M</td>
<td>0.0%</td>
<td>0.0%</td>
<td>4(3.9%)</td>
<td>8(7.7%)</td>
</tr>
<tr>
<td></td>
<td>F</td>
<td>4(3.9%)</td>
<td>4(3.9%)</td>
<td>16(15.4%)</td>
<td>44(42.3%)</td>
</tr>
</tbody>
</table>

Males: $X^2_{0.05,6} = 12.592 \text{ (P or Book value)}$, Calculated Value = 8.212 (P> 0.05), Females: Calculated value = 13.577 (P< 0.05)
Discussion

The ABO locus is associated with the serum levels of molecules known to bind to *P. falciparum* infected red blood cells that are also markers of damage to vascular endothelial cells and inflammatory processes (Barbalic *et al*., 2010; Cserti-Gazdewich *et al*., 2011; Paterson *et al*., 2009). Furthermore, parasitized red blood cells have a stronger tendency to form rosettes with uninfected erythrocytes of the A, B or AB blood groups than with those of blood groups O (Barragan *et al*., 2000). In an earlier study, Fischer and Boone (1998) reported that it is not clear whether blood groups via their influence on rosette formation are causally associated with severe malaria or whether they merely serve as markers for other host-parasite interactions that provoke the severe manifestations of malaria. Some researchers have reported that haemoglobin genotype is a factor in susceptibility to plasmodium species infection in humans (Hill *et al*., 1992; Ademowo *et al*., 1995; Omotade *et al*., 1999). But there is lack of consensus on possible association between ABO blood group genes and malaria parasitaemia Hill *et al*., 1992; Ademowo *et al*., 1995; Omotade *et al*., 1999; Fischer and Boone, 1998).

A number of studies have been conducted to investigate the association between ABO blood group system and some disease conditions (Zerihun *et al*., 2011). Some of these studies reported significant associations suggesting that ABO blood groups have an impact on infection status of the individuals possessing a particular ABO blood group (Beiguelman *et al*., 2003; Santos *et al*., 1983). Some studies reported the absence of significant association between *P.*
Results obtained in this study showed that 4(3.9%), 16(15.4%), 32(30.8%) and 52(50.0%) of the 104 made up of 24(23.1%) male and 80(76.8%) female students screened, belonged to blood groups AB, A, and O respectively in that increasing order. These findings showed that the most prevalent blood group is O(50.0%) and the least, AB (3.9%). This was consistent with the report of Ilozumba and Uzozie (2009) which stated blood group prevalence rates of 2.63%, 12.05%, 21.0% and 63.83% for AB, B, A and O respectively. Results in this study also differed from that of Otajevwo (1997) which reported prevalence rates of Warri ABO blood groups of 12(6.9%), 33(19.0%), 36(20.7%) and 93(53.5%) for AB, B, A and O respectively. Findings in this work also differed from the report of Zerihun et al. (2011) which recorded 3.3%, 21.9%, 23.5% and 51.3% for AB, B, A and O respectively.

Data obtained in this work also indicated that there were 24(23.1%) and 76(73.0%) male and female rhesus positive students respectively and whereas, there were no male rhesus negative individuals, a total of 4(3.9%) female rhesus negative students were recorded (Table 1). This confirmed that although the prevalence of rhesus negative factor in populations is small, the occurrence of 4(3.9%) out of a sample population of 104 may not be insignificant in view of the medical implications in terms of child birth and still birth.
Data obtained in this study also showed that the most prevalent blood group is group O because 52 students representing exactly 50.0% of the population sampled, belonged to that group. At the other extreme, the least occurring group was AB with 4(3.9%) students occurring in the group (Table 2). These findings are not consistent with an earlier study by Agbonlabor et al. (1993) which reported groups O (51.9%) and A (11.0%) as the highest and least occurring respectively, but are in agreement with the reports of Zerihun et al. (2011), Ilozumba and Uzoekie (2009) and Otajevwo (1997).

Results further revealed that 80(76.8%) of the 104 students harboured malaria parasites of various degrees of density. This rather high parasitaemia rate suggests that Okada town is hyperendemic for malaria. This parasitaemia rate appears to be low when compared to 93.4% obtained in Odoakpu, Onitsha South by Ilozumba and Uzoekie (2009). Parasitaemia rate in this study appears high however, when related to prevalence rate of 79.3%, 77.4%, 58.3%, 43.2%, 10.0% and 6.0% obtained respectively for blood donors in Warri (Otajevwo, 1997) Owerri (Mbanugo and Emenalo, 2004), Children in Awka (Mbanugo and Ejims, 2000), coastal dwellers of Lagos State (Nebe et al., 2002), blood donors in Ibadan (Edington and Gillies, 1976) and blood donors in Mainduguri (Ahmed et al., 2001). These results may suggest existence of regional differences in malaria parasitaemia in Nigeria with the eastern area (represented by Onitsha South LGA and Owerri) ranking highest in prevalence rating and the Northern area (represented by Maiduguri) occupying the lowest position, while the Western area
(represented by Lagos State and Ibadan takes a middle position Ilozumba and Uzozie, 2009). Results of more studies on malaria parasitaemia in different parts of the country would be needed before a more definite statement on the apparent trend could be made.

The somewhat high malaria parasitaemia rate recorded among Igbinedion University, Okada students in this study may be as a result of the unsanitary situation of the University environment (with weeds and unkept grass around classrooms) or the unsanitary situation in and around the students’ hostels in terms of choked drainage channels, littering of empty cans, collection of pools of water etc which provide breeding grounds for anopheles mosquitoes (vectors of human Plasmodium species) which breed in diverse types of aquatic habitats (Chandler and Read, 1961). Other likely pre-disposing factors in the area may include overcrowding in hostels which could facilitate vector-man contact, and malaria parasite transmission.

Out of 24(23.1%) male students screened 12(11.5%) were infected compared to 68(65.4%) female infected blood samples out of a total of 80 (76.8%) female samples processed. This clearly shows a much higher infection rate in females in relation to the males this reports is not consistent with trends observed in Warri (Otajevwo, 1997), Ekpoma (Agbonlahor et al., 1993), Odoakpu (Ilozumba and Uzozie, 2009), Owerri (Mbanugo and Emenalo, 2004), coastal areas of Lagos (Nebe et al., 2002; Afolabi et al., 1997). The reasons for the
observed gender differences are far-fetched and more studies of other populations in other of Nigeria would be expedient to offer possible explanation.

Data obtained in this study also showed that the highest malaria parasitaemia rate was observed in the O blood group with 40(38.4%) group O students showing various densities of *Plasmodium falciparum* parasites out of 52(50.0%) group O students screened. Again, this finding is in agreement with those of Otajevwo (1997) and Nkuo-Akenji (2004). Ilozumba and Uzozie (2009) recorded 100.0% malaria parasitaemia among blood group AB individuals followed by 94.9% among blood group O subjects and this is not consistent with present study. Findings in present study also disagree with Migot-Nabias *et al.* (2000) and Pathirana *et al.* (2005) who observed low parasitaemia among blood group O individuals. In their reports, they concluded that blood group O seems to confer a certain degree of protection against severe courses of the disease.

It is note-worthy that blood group A subjects appeared to record a high parasitaemia rate of 12(11.5%) out of a total of 16(15.4%) who belonged to the group. Parasitaemia rates of 24(23.1%) out of 32(30.8%) and 4(3.9%) out of 4(3.9%) for blood groups B and AB also appear to be high relative to each other. Although a study using larger sample size would be required to validate this finding, it seems there is relative spread of malaria parasitaemia across all blood groups in this study. This is consistent with findings from a study carried by Fischer and Boone (1998) of which they reported that malaria occurs in patients of
any blood group and that no particular blood group precludes the possibility of severe malaria.

An equal number of male and female students occurred in blood group A. In group B, 28(26.9%) female students occurred compared to 4(3.9%) male students. This somewhat suggests that there are more females in group B when compared with male blood group B individuals. Whereas there were no male AB individuals, all the 4(3.9%) screened were female and ostensibly maintaining the earlier trends (apart from group A), more than three-fold female students occurred in group O compared with the males. The gender discriminatory trend seems to erroneously suggest that more female individuals in Igbinedion University, Okada occur in each blood group. The only plausible reason adducible is that it is by chance. The picture may become clearer more acceptable if a similar study with a much higher sample size is carried out possibly using the same study area.

Also going by sex distribution of malaria parasitaemia, results revealed that much more female students were infected with malaria parasites compared to the male students. This does not tally with the suggestion of Portilo and Sullivan (1979) that genetic factors could play a role by endowing females with immuno-regulatory potentials to cope better with some diseases. Again, the occurrence of much more malaria parasitaemia rate among the female students across all blood groups (Table 3) may be a matter of chance. This, too, is subject to experimental re-evaluation by some other researchers.
There was a significant statistical association between malaria parasitaemia and ABO blood groups ($X^2_{0.05,6} = 12.592$, calculated value at 95% C.I = 13.663 and hence, $P < 0.05$). This significant association is in line with the report of Zerihun et al. (2011) and is inconsistent with findings of Nkou-Akenji (2004), Martin and Miller (1979), Facer and Brown (1979), Kassim et al. (1982), Bayoumi et al. (1986), Akinboye and Ogunrinade (1987), Thakur and Verma (1992), Montoya et al. (1994) and Uneke et al. (2006). There was also statistical significant association of malaria parasitaemia and ABO blood groups among female students ($X^2_{0.05,6} = 12.592$, calculated value at 95% C.I = 13.577 and hence, $P < 0.05$). The association of malaria parasitaemia with ABO blood groups among male students was not significant ($X^2_{0.05,6} = 12.592$, calculated value at 95% C.I = 8.212 and $P > 0.05$). Despite these results, it is still difficult to make a clear conclusion on the association of ABO blood groups and malaria parasitaemia because a lot of study designs have been conducted invivo.
REFERENCES


44. Nail, SJ; Mc knight A, Gustafsson K, and weiss RA (2005) HIV – 1 incorporates ABO histo - blood group antigen that sanitizer visions to complement mediated in activation. *Blood* 105 (12); 4693-9.


54. Santos SEB, Salzano Fm, Franco MHLP, and Ereitas MJM (1983) mobility, genetic markers susceptibility to malaria and race mixture in manaus, Brazil, J hum Evol 12: 373-381.


