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ABO and Rh D Blood Groups in *Plasmodium falciparum* Infected Subjects at Aminu Kano Teaching Hospital, Kano-Nigeria

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Abstract

The interplay between malaria parasites and blood group antigens remains a fascinating subject with potential to contribute to the development of new interventions to reduce the global burden of malaria. Elucidation of the ABO and Rh D blood group antigens status and infection with Plasmodium falciparum can bring about understanding of the differences noted in the ABO blood group variation in malaria endemic areas. The aim of this study was to assess the distribution of ABO and Rh D blood groups in relation to P. falciparum malaria infection in subjects infected with malaria receiving medical care at Aminu Kano Teaching Hospital, Kano. A total of 388 plasmodium falciparum infected subjects were recruited for this study. The ABO and Rh D blood groups of all participants were determined using standard tube technique while P. falciparum infection and the parasitaemia levels were also determined microscopically by thin and thick blood films, respectively. The blood groups were O Rh D+ 190 (48.97%), blood group A Rh D+ 95 (24.48%), blood group B Rh D+ 83 (21.39%), blood group AB^+ 14(3.61%), blood group O Rh D- 3 (0.77%), blood group A RhD- 1 (0.26%), blood group B Rh D- 2 (0.52%), and blood group AB Rh D- 0 (0%). The result of this research shows that the ABO blood group O is the most common, followed by group A, group B is less common and group AB is the least common. We concluded that blood group O offers a survival advantage, group A offers a disadvantage, and group B has an intermediate effect.

Keywords: Antigens, ABO blood group, Rh factor, Plasmodium falciparum, Malaria.

Introduction

Malaria is a life-threatening blood borne disease caused by Plasmodium parasites transmitted to humans by the infected female *Anopheles* mosquito.

Malaria is a protozoan disease that has infects humans for over 50,000 years, and *Plasmodium* may have been a human pathogen for the entire history of the species (Joy et al., 2003). The disease is caused by various etiological agents including Plasmodium falciparum, Plasmodium malariae, Plasmodium ovale, Plasmodium vivax and Plasmodium knowlesi. While *P. vivax* is responsible for the largest number of malaria infections worldwide, P. falciparum accounts for about 90% of deaths from malaria (Mendis et al... 2001). Ninety percent of deaths occurred in Sub-Saharan Africa where *Plasmodium falciparum* is the most prevalent specie and the leading cause of malaria deaths (World Malaria Report, 2014). P. falciparum has been called "the strongest known force for evolutionary selection in the recent history of the human genome" (Kwiatkowski, 2005). One such gene that is selected for by malaria is the ABO blood group gene which has three alleles namely, A, B and O, coding for different types of agglutinogens attached to the surface of red blood cells (RBCs) and hence determining an individual's blood group.

The World Health Organization estimated that there are 250 million cases of fever annually due to malaria (WHO, 2009). In 2010, WHO estimated that 655,000 people died from Malaria (World Malaria Report, 2011). However, according to studies done at the University of Washington and University of Queensland estimated significantly higher deaths records, with 1,238,000 people dying from malaria in 2010 (Murray et al., 2012). Despite the high morbidity and mortality, certain individuals are resistant to malaria infection due to different immune responses by the host and to a varying extent, and certain characteristics possessing protective value against infection such as glucose 6 phosphate dehydrogenase deficiency, sickle cell trait (HbAS) and sickle cell disease (HbS) (Otajevwo, 2013).

The plasmodium parasite has established a close relationship between itself and the RBCs. Indeed, the severe pathophysiological manifestations of malaria caused by *Plasmodium falciparum* are direct consequences of the parasite's blood stage replication cycle, as *merozoites* repeatedly invade, multiply within, and destroy RBCs. Consequently, RBCs have evolved specific receptor-ligand interactions, some of which involve the ABO blood group antigens, to facilitate their adherence and invasion by *merozoites*. Therefore, any variation in the erythrocyte ABO antigens can change the penetration and establishment of the parasite in the merozoites (Simiyu, 2013).

There is increasing evidence that *P. falciparum* malaria is influenced by ABO blood type of an individual but the extent of association is not fully established (Ilozumba and Uzozie, 2009). However, several studies undertaken have been unable to link ABO blood groups to the incidence of malaria or to the repeat attacks of malaria. Recent studies on pathogenesis of malaria have shown that parasite triggered red blood cell rosette formation in association with the severity of clinical manifestation and malaria. In this study, we aimed to investigate the relationship of *Plasmodium falciparum* infection and blood groups antigens.

Materials and methods Subjects

The study was a prospective cross-sectional study on *Plasmodium falciparum* infected subjects of all ages and gender receiving medical care at Aminu Kano Teaching Hospital, Kano.

Ethical considerations

Blood samples were obtained from consenting subjects who were enrolled into the study, in accordance with institutional guidelines set forth by Aminu Kano Teaching Hospital (AKTH) as per declaration of Helsinki ethical principles for medical research involving human subjects.

Sample collection and processing

Five milliliters of venous blood were collected from

Table 1: Characteristics of the study subjects

Gender	Number (n)	Percentage (%)
Males (%)	190	(48.97)
Females (%)	198	(51.03)
Total	388	(100)
Age (M±SD)	20.7 ± 16.1	

each study participant in EDTA container for ABO/Rh D blood grouping and blood film preparation. ABO and Rh D blood grouping was done using commercially prepared monoclonal anti-A, anti-B, and anti-D antisera based on the method of Cheesbrough, (2010) and its modifications. Thick and thin blood films were prepared and stained appropriately for malaria parasitemia and species identification with aid of microscope, respectively.

Statistical analysis

The results were analyzed using Graph pad prism statistical tool (version 7.2) and the results expressed as frequencies, percentages and mean plus or minus standard deviation. And the correlation was compared. Ap-value of ≤ 0.05 was considered as significant in all statistical analysis.

Results

A total of 388 plasmodium falciparum infected subjects, comprising of 190 (48.97%) and 198 (51.03%) males and females, respectively. The mean and standard deviation of age of the study population was 20.7 ± 16.1 (Table 1). In (Table 2), the patterns of ABO and Rh D blood groups were presented by gender. The frequency of ABO blood group was in the following order O > A > B > AB in both male and females. Table 3 shows the distribution of ABO blood group of all subjects with blood group A = 96(24.7%), B = 85(21.9%), AB = 14(3.6%), and O = 193(49.8%), respectively.

The parasitemia level in different ABO blood group (A, B, AB, and O) is present in table 4. Parasitemia level was in the following order B > A > O and > AB, respectively. There was a statistically significant (p< 0.001) difference in the level of parasitemia in the different ABO blood groups. Table 5 shows the comparison between the level of parasitaemia based on gender of subjects. The result shows no statistically significant difference between the parasitaemia level and gender. Table 6 shows the distribution of Rh D group among the study participants. Three hundred and eighty- three (98.45%) of the study participants were Rh D positive and six (1.55%) were Rh D negative.

Key:

n = Number of subjects

M=Mean

SD= Standard deviation

Table 2: Pattern of ABO and Rh D blood groups of plasmodium falciparum infected subjects by gender

Blood groups	Males (n=190)	percentage %	Females (n=198)	Percentage %
A+	45	(23.68)	50	(25.25)
B+	37	(19.47)	46	(23.23)
AB+	7	(3.68)	7	(3.53)
O+	99	(52.11)	91	(45.96)
A-	0	(0)	1	(0.51)
B-	1	(0.53)	1	(0.51)
AB-	0	(0)	0	(0)
O-	1	(0.53)	2	(1.01)

Table 3 Distribution of different ABO and Rh blood groups among the study subjects

Blood groups	Subjects (n)	Percentage (%)
A+	95	(24.48)
B+	83	(21.39)
AB+	14	(3.61)
O+	190	(48.97)
A-	1	(0.26)
B -	2	(0.52)
AB-	0	(0)
O-	3	(0.77)
Total	388	(100)

Table 4 Parasitemia level in different ABO blood group

Blood groups	Subjects	percentage %	Parasitaemia Level (μl ⁻¹ of Blood)
A+	95	(24.48)	2842.1 ± 1242.6*
B+	83	(21.39)	$2114.5 \pm 884.57*$
AB+	14	(3.61)	$2071.4 \pm 583.66*$
O+	190	(48.97)	$1565.8 \pm 804.84*$
В-	2	(0.52)	2750.0 ± 1060.7 *
O -	3	(0.77)	$1166.7 \pm 577.35*$

(Mean \pm SD) *p<0.0001- statistically significant difference

Table 5 Comparison of Parasitaemia level and gender

Blood	Subjects	Mean parasitaemia $\mu\Gamma^1$ of blood (Mean \pm SD)	
groups	(n=388)		
Males (%)	190 (48.97)	2000.0±1056.6**	
Females (%)	198(51.03)	2055.6±1105.8**	

Key:

n = Number of subjects

SD= Standard deviation

Table 6 Distribution of Rh factor among the study population

Rh D	Number of malaria infected subjects	
Group	n (%)	
Positive	382 (98.45%)	
Negative	6 (1.55%)	
Total	388 (100%)	

Key:

n = Number of subjects

Discussion

In this present study, The ABO blood group of the study population indicates that group O is the most common, followed by group A, group B and group AB is the least common. However, with respect to Rh blood group, O Rh negative was highest followed by group B negative and A negative being the least frequent, respectively. Therefore, our study indicates that blood group O positive is the predominant and blood group A negative was the least and AB negative was the rarest. This is concurrence with the reports from studies done in North-Eastern Nigeria (Tidi et al., 2013; Ilozumba et al., 2009) in Anambra state. However, our finding is at variance with a previous report (Tewodros et al., 2011). These differences may be due to ethnic and geographical locations of the study populations.

Results from this study indicates comparison of mean parasitaemia level of individuals with blood groups A positive, B positive, AB positive and B negative were statistically significant (p <0.0001). Whereas, comparison of mean parasitaemia between blood groups O positive and O negative were statistically not significant (p>0.05). Our findings is in conformity with study done by Barragan *et al.* (2000) who reported that blood group A is a risk factor for severe malaria, whereas blood group O may offer some protection against severity of the disease. The findings from this study is also in agreement with a

study done in Gabon by Migot-Nabias *et al.* (2000) and that of Pathirana *et al.* (2005) who observed a low *P. falciparum* parasitaemia among blood group O individuals. In their reports, they concluded that blood group O seemed to confer a certain degree of protection against severe courses of the disease.

Furthermore, in similar studies done in Gambia a protection role in blood group O was observed against severe malaria. Varying degrees of susceptibility to P. falciparum malaria among individuals with blood group A and B and resistance to P. falciparum malaria to P. falciparum malaria by blood group O have been reported (Carton et al., 1983). This may indicate that individuals with these blood groups are more prone to severe *P. falciparum* infection than the other blood groups. This still stand to mean that an individual's blood group may be a risk factor to predisposition to P. falciparum infection. Therefore, the differences between the mean parasitaemia levels per ul of blood are not likely to be due to chance but rather due to blood type the individual possess. This stressed the relationship between P. falciparum parasitaemia level and the ABO blood group type.

We also documented here, that both male and female subjects were equally at risk of getting infected. However, disease prevalence was slightly higher in females than males (p>0.05). This finding is in

^{**}p>0.6134 – no statistically significant difference

agreement with a study done in Edo state Nigeria (Otajevwo 2013). However, this is also contrasted with the findings of a study done in Ondo state, Nigeria by Simon-oke *et al.* (2016) who reported males had a higher malaria parasite infection rate than females.

Considering the mean parasitaemia levels for gender, this study reveals that there was no statistical significance difference based on gender (p>0.6134). This is in contrast with the finding by Otajevwo (1997) in a study associating malaria parasitaemia with ABO blood groups among residents of Warri, Nigeria which indicatedthat malaria parasitaemia was higher among the males than females. Similarly, a previous report by Uzoegwu and Onourah (2003) reported similar results that males has higher parasitaemia than females.

Considering mean parasitaemia levels in individuals of different age groups, it was observed that age group 0-9 have high levels of mean parasitaemia followed by individuals of age group 60-69 year. We hypothesize that this finding may be due to the low immune competence of individuals within that age ranges. However, this is consistent with the findings of some studies done in Edo State, Niger-Delta Region of Nigeria by Onyesom *and* Onyemakonor (2011) and that of Ngonga (2012) in Kenya.

Our present study documented that majority of our study population were Rh positive and very few were Rh negative. This is in agreement with a study done in Niger-Delta region of Nigeria by Emelike *et al.* (2010) who reported 91.3 % and 8.7% for Rh positive and Rh negative respectively. This can be compared with a study done on the distribution of ABO and Rh blood groups among indigenes of Abuja, Nigeria by Olaniyan *et al*, (2013) who reported a high prevalence of Rh positive subjects compared to Rh negative subjects 95.7 % and 4.3 %, respectively.

Conclusion

The findings of our present study showed that blood group O positive is less vulnerable to *P. falciparum* infection among the study population. It also established a significant relationship between the ABO and Rh blood groups with P. *falciparum* malaria infection, where individuals of blood group O positive were found with the least parasitaemia level, individuals of blood group A positive were found to have high parasitaemia level. Blood group B appeared to confer an intermediate effect. The findings of this study therefore agreed with the proposed hypothesis that during an infection with *P*.

falciparum, blood group O offers a survival advantage, group A offers a disadvantage, and group B has an intermediate effect.

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Conflict of Interest:

Conflicting interest none.

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