The Prevalence of Malaria Parasitaemia and Predisposition of ABO Blood Groups to *Plasmodium falciparum* Malaria among Blood Donors at a Ghanaian Hospital

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Abstract

There is increasing evidence that Plasmodium falciparum *malaria is influenced by ABO blood group but the extent of association between the two is yet to be well defined. This study was sought to determine if certain individuals are predisposed to* P. falciparum *malaria parasite infection by virtue of their ABO blood group, and the malaria parasitaemia prevalence rate among presumably healthy donors. A total of 437 blood samples were examined following best practices. Thick and thin blood films were made from each sample and ABO blood groups determined by a standard tube agglutination technique. Of the 437 samples examined, 13% had malaria parasite in their blood, all of which was identified as* P. falcipurum. *Most of the donors, 70.5% (308) were females, while 29.5% (129) were males. Of the 129 male donors, 19.4% had malaria parasites in their blood, while 10.4% of the 308 females had malaria parasites. The infection percentage was significantly higher in the males compared to the females. The respective infective rates were 14.3, 11.1, 13.9 and 0.00% for the blood groups A, B, O and AB. The difference in infection percentage between the various blood groups was, however, not statistically significant. The malaria prevalence rate of 13.0% among the donors in this study is an indication of asymptomatic parasitaemia among blood donors and thus poses a great risk to blood recipients.*

Keywords: *Transfusion, thick and thin blood films, standard tube agglutination technique, blood recipients.*

1. Introduction

Malaria is a very important disease in Sub-Saharan Africa with high morbidity and mortality rates (WHO 1994), which consequently leads to a dwindle in the productivity of a nation (WHO 1994). The most common individuals at high risk of malaria infection in endemic areas are people of low immunity, for instance, foreigners, pregnant women, children (Weir and Stewart 1997) and perhaps HIV/AIDS patients (Migot *et al*. 1996; Chandramohan and Greenwood 1998).

Transfusion therapy is the use of blood and blood products as a form of treatment to save lives. However, this process can lead to transfusion transmitted infections such as malaria parasites if blood is received from a malaria infected donor (Okocha *et al*. 2005). An initial report on malaria as a result of blood transfusion has been published in 1911 (Woolsey 1911). Evidence shows that all the four human parasites of malaria, which are *P. falciparum, P. malariae, P. vivax* and *P. Ovale*, may possibly be transmitted through blood transfusion (Wylie 1993). Of outmost concern is that the majority of the individuals that require blood transfusion are often already destabilised by one disease or the other and their immunity is already compromised. The malaria parasites behave violently in these individuals leading to very serious complications and even

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death (Ali *et al*. 2004). It has also been evidenced that the human plasmodium parasite can continue to exist in stored and frozen blood (Talib and Khurana 1995), which makes malaria transmission by blood transfusion even more dangerous and of greater concern.

The WHO reports that, there are about 300-500 million incidences of malaria causing 2-3 million deaths each year in the tropical and subtropical regions of the world. About 90% of these deaths occur in Sub-Saharan Africa (WHO 2003). This could be as a result of *P. falciparum* which is the most dangerous of the four parasites being the main cause of infections in this region and also the main malaria transmitting vector (*Anopheles gambiae*) being spread widely and very difficult to control (WHO 2003).

Regardless of the soaring malaria incidence in endemic regions, a certain group of individuals seem to have more immunity to malaria than others. This could be accounted by several factors including haemoglobin variants, ABO blood group system and enzyme action, among others (Otajevwo 2013). An investigation into malaria in correlation with, for instance, ABO blood group has the potential of giving an insight into the pathogenesis of malaria and perhaps aid the control of this disease.

The link between ABO blood groups and the incidence of malaria parasitaemia or immunity to malaria is still unclear (Akinboye and Ogunrinade 1987; Thakur and Verma 1992). This is probably because the relations between the blood group and malaria have not been well studied (Singh *et al*. 1995). Other studies have revealed that the formation of rosette which is triggered by malaria parasite is linked to the rigorousness of clinical disease (Rowe *et al*. 1995) as well as cerebral malaria (Treutiger *et al*. 1992). Interestingly, some researchers have found that, these rosettes are formed better depending on the blood cell types, with the blood cell type A and B having higher chances of forming rosettes (Carlson and Wahlgren 1992; Udomsangpetch *et al*. 1993). Other studies have also shown that blood types A, B and AB are more vulnerable to plasmodium falciparum malaria than the O blood type (Omotade *et al*. 1999), whereas others have reported an equal vulnerability among the various ABO blood types (Otajevwo 2013). There is, therefore, no consensus yet, hence the need for more research in this respect to help the understanding of malaria pathogenesis with regards to its association with ABO blood type.

This study was aimed at determining the prevalence of malaria parasitaemia among the blood donors at Komfo Anokye Teaching Hospital in Ghana and also to investigate if certain individuals by virtue of their ABO blood type are predisposed to getting malaria.

2. Materials and Methods

2.1 Ethical Clearance

This study was conducted at the blood bank of the Komfo Anokye Teaching Hospital (KATH), Kumasi, Ghana. The hospital serves as a referral centre for blood transfusion services in the Ashanti region. The relevant approval for this study was obtained from the committee of human research publications and ethics at KATH and Kwame Nkrumah University of Science and Technology, Kumasi.

2.2 Inclusion and Exclusion Criteria

A total of 437 apparently healthy blood donors were considered for the study. Subjects were made up of males and females. The donors included volunteer and replacement blood donors, with blood pressures between 90-140/60-90 mmHg, body weight of 50 kg and above. Each donor was also screened for HbsAg, HCV, and HIV-antibodies. Their Hb values were not less than 12 g/dl or 13 g/dl for females and males, respectively. Excluded from the study were those taking drugs for high blood pressure, having Hb below the above, who have tested positive for HbsAg, HCV, or HIV-antibodies, who have had jaundice, liver disease, epilepsy, diabetes, duodenal or gastric ulcer, asthma and tuberculosis, or sickle cell disease. Prostitutes, homosexuals, drug addicts, those taking self-injected drugs, and those who weighed less than 50 kg were also excluded.

2.3 Phlebotomy and Laboratory Analysis

Blood samples were collected after swabbing the area to be sampled with 70% alcohol and air dried. Venous blood was collected from the median cubital vein of the

study subjects using 5-ml capacity disposable syringes fitted with needles. The blood was dispensed into specimen containers containing pre-measured amount of dipotasium salt of ethylene diamine tetra acetic acid (EDTA). Gentle mixing of the EDTA and the blood was ensured and then thick and thin blood films were prepared from the venous blood containing EDTA. The films were stained for 30 minutes in 5% solution of Giemsa stain. Blood films were examined under ×100 objective lens (immersion oil) using a light microscope as described by the WHO method (WHO 1993).

Parasite densities were calculated using the thick films by the WHO method (parasite count × 6,000 divided by the number of WBCs counted) (WHO 1993), and the thin films were used to establish the species and infective stages of the parasites. Films were classed as negative when no parasites were seen after two hundred microscopic fields were being examined. For purposes of quality control, slides were cross-examined independently by a senior biomedical scientist and the results compared. The patients' various ABO blood groups were also determined by a standard tube agglutination technique (forward and reverse ABO blood grouping) (WHO 1993).

2.4 Statistical Analysis

The results are presented as Mean ± SD. All categorical variables were analysed using Chi-Square Test. Statistical analysis was performed using GraphPad Prism software and significance achieved when $P < 0.05$.

3. Results

The demographic characteristics of the donors screened in this study are summarized in Table 1. The mean age of the donors was 17.8 ± 3.3 (mean ± SD) with a range of 15-50 yrs. Most of the donors 70.5% (308) were females, while 29.5% (129) were males. The majority of the donors (90.6%) were aged 15-20, followed by 21-25 (6.2%) and then 1.6% for both 26-30 and >30 age groups.

The prevalence of the various ABO blood groups were 20.82, 18.54, 57.44 and 3.20% for Groups A, B, O and AB, respectively. About 97.71% of the donors were rh D positive, while only 2.29% of them were rh D negative.

The distribution of the various blood groups in relation to sex is summarized in Table 2. The majority of the donors among both males and females belonged to group O and there was no statistically significant difference in the occurrence of ABO blood group phenotype in males and females ($P = 0.3098$).

Of the 129 male donors examined, about 19.4% had malaria parasites in their blood, while 10.7% of the 308 females had malaria parasites. The infection percentage was significantly higher among the males compared to the females ($P = 0.0199$) as shown in Fig. 1. The odds of developing malaria infection among the males were 2.003 relative to the females (OR = 2.003; 95% CI = 1.137 to 3.530). All the malaria infections were identified as *P. falciparum* and of the 437 total samples examined, 57 (13%) had malaria parasites in their blood.

Table 1. Demographic characteristic of the study subjects.

Parameter	*N* (%)
Age	
15-20	396(90.6)
21-25	27(6.2)
26-30	7(1.6)
>30	7(1.6)
ABO	
A	91(20.82)
B	81(18.52)
O	251(57.44)
AB	14(3.20)
Rh factor	
Rh positive	427(97.70)
Rh negative	10(2.29)
Sex	
Male	129(29.5)
Female	308(70.5)

Table 2. Distribution of ABO blood groups by sex.

Sex	A	B	AB	O
Male	31	20	0	78
Female	60	61	14	173

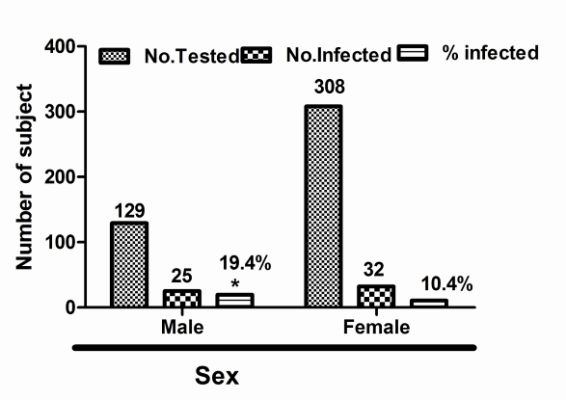

Fig. 1. The prevalence of malaria parasitaemia among male and female donors.

The prevalence of malaria parasites in blood donors according to age is shown in Table 3. The age group of 21-25 years recorded the highest prevalence rate (22%) and mean parasite density of 2,000 parasites/µl of blood, followed by the age group of >30 years of 14% prevalence rate and 2,000 mean parasite density. The age group of 15-20 years had a prevalence rate of 13% with the highest mean parasite density of 2,234.69/µl (Table 3).

The respective infective rate and the mean parasite densities are 14.3% and 1307.69/µL, 11.1% and 2,055.56/µL, 13.9% and 2571.43/µL and 0.0 for blood groups A, B, O and AB as shown in Table 4. The difference in infection percentage between the various blood groups was, however, not statistically significant ($\chi^2 = 5.528$; $P = 0.1370$).

Table 3. Prevalence of malaria parasites in donors according to age.

Age	Examination number	Infection percentage	Mean parasite density/µL
15-20	396	13	22,234.69
21-25	27	22	2,000.00
26-30	7	0	0.00
>30	7	14	2,000.00

Table 4. Prevalence of Malaria parasitaemia among the ABO blood groups.

ABO blood group	Examination number	Infection percentage	Mean parasite density/µL
A	91	14.3	1,307.69
B	81	11.1	2,055.56
AB	14	0.0	0.0
O	251	13.9	2,571.43

4. Discussion

Safe blood transfusion has been a key worry to health workers since the beginning of blood transfusion many years ago. The malaria prevalence rate of 13.0% among blood samples examined in this study, though not very high, still poses some concerns. In fact, these were donors that were presumed to be healthy and perhaps reflect the high endemicity of malaria in the country. The implication of this is that thirteen in one hundred blood donors bear the risk of passing on malaria parasites to their recipients. Unfortunately, the most common group of patients who need blood transfusion fall under the malaria vulnerable group (e.g. pregnant women, children under five years and the immunocompromised patients) (Qari *et al*. 1993). If these groups of people are transfused with malaria infected blood, it could exacerbate their conditions.

The prevalence rate in this study was, however, low compared to 40.9% by Uneke (2007) among donors in South-Eastern Nigeria, 30.2% by Okocha *et al*. (2005) in Nnewi, Nigeria, and 51.5% by Epidi *et al*. (2008) among blood donors in Abakaliki in Nigeria. The low rate in the current study could be due to the difference in the study populations.

It has been evidenced that in countries where blood donation is a commercial business deal, transfusion transmitted malaria is very common (Enosolease *et al*. 2004).

The issue of commercial donors has not gained grounds in Ghana, and Kumasi in particular, where the majority of the donors are from the various senior high schools and blood donation clubs in the Kumasi metropolis, thus perhaps another reason for the comparatively low rate in this study.

The infection percentage was significantly higher in males than females, 19.4% vs. 10.7% for males and females, respectively. The odds of developing malaria were about twice higher in males relative to females. The finding in this study is similar to the work of Bonilla and Rodriguez (1993), where males had a higher malaria parasite infection rate than females. The presents study was, however, in contrast to that of Vlassoff and Bonilla (1994) which reported more females being infected than males, and also that of Otajevwo (2013) which reported a higher infection rate in females than males. One interesting feature in this study was the fact that there were more female donors than males and this was encouraging because blood donation was originally seen as the preserve of the male gender in Ghana. The low rate of infection among female donors also goes to suggest that female students were less exposed to mosquitoes that transmit the *P. falcipurm* malaria parasite.

Interestingly, in this study, the mean parasite densities decreased with an increase in age and although individuals aged between 15-20 years had the highest density of 2,234.69/µl, individuals with age between 21-25 years had the highest infection rate of 22.0%. The reason for this difference observed in terms of age was not actually clear and needs further investigation. It was, however, clear that the majority of the infected individuals were the young adults and the adolescence. Since these groups of people are the principal blood donors (WHO 2003), there is therefore a strong need to intensify malaria control programs among these groups.

The infection percentage rate in a decreasing order was as follows: blood group A, followed by O, then B, and AB being the least infected, even though the difference was not statistically significant. This findings is similar to the studies of Uneke (2007) in South-Eastern Nigeria who found that there was no significant relationship between ABO blood groups and *P. falciparum* malaria, and also similar to the work of Otajevwo (2013) who found that any of the ABO blood groups were at equal chances of malaria infection. However, it contradicts the study of Singh *et al*. (1995) who reported that group B was the most vulnerable (41.8%), followed by group A (29%), then group O (22.2%), and group AB being the least vulnerable (7%).

5. Conclusion

The malaria prevalence rate of 13.0% among the donors in this study is an indication of asymptomatic parasitaemia among blood donors and thus poses a great risk to blood recipients, especially the malaria vulnerable groups.

The findings of this study also did not support the assertion that individuals with certain blood groups were predisposed to *Plasmodium falciparum* malaria infection. This implies that there should not be any bias towards a particular ABO blood type in the management and control of malaria.

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