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Malaria Parasitemia: It's Association with Bacteramia, Haemoglobin Genotype and ABO Blood Group System

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Abstract: The aim of this study was to investigate the distribution of malaria parasitemia among suspected cases of malaria, with the ABO blood group and haemoglobin genotype and also to find out if bacteramia is a normal concurrent infection in malarious patients. Five hundred and fifteen suspected cases were examined by Giemsa staining method using both thick and thin blood films. The simple code from one to four crosses was used to determine relative malaria parasite count. Four hundred and fifteen (80.6%) were positive for malaria parasite. The prevalence was 222 (89.2%), 193 (72.6%) for females and males, respectively. There was high prevalence of malaria parasitemia in blood group O (90.0%) and lowest was in blood group AB (31.3%). Considering haemoglobin genotype, 311 (85.2%) was the prevalence for malaria parasite in AA, with genotype SS recording the least with (44.0%). Of all the 515 cases examined in this study, bacteria were isolated from the blood of only eight subjects (1.6%) and this was considered insignificant ($p>0.05$) thus, pointing out that bacteramia may not be a usual occurrence with malaria parasitemia. The age group 1-10 years had the highest occurrence of parasitemia with (91.7%) while the least was obtained in age range 21-30 years with (70.9%). It was observed that the density of malaria parasitemia was highest in age group 1-10 years meaning parasite density decreases with increasing age. Also, genotype AA had the highest malaria density.

Key words: Prevalence, malaria parasitemia, ABO blood group, haemoglobin genotype, bacteramia

INTRODUCTION

Malaria is still one of the greatest public health problems in many tropical and subtropical countries (Uneke, 2006). It may also occur in temperate region with its dissemination diminishing from the equator (Biezle *et al*., 1980; Werner and Friedrich, 1987). Malaria is a disease or condition of infection in man caused by intracellular protozoan parasites belonging to the genus plasmodium (Uneke, 2006). Mortality and morbidity through anaemia, cerebral complications or other mechanisms are mainly associated with Plasmodium falciparum (Uneke, 2006). Of the 300-500 m clinical cases of malaria reported annually more than 90 million are from Africa (Isibor *et al*., 2003). An estimated 280 million people are carriers of the malaria parasite in the region (Mahmoud, 1987; Benzerrong and Elo, 1991).

According to a United Nations Population Division report (1990), malaria is the only disease today, apart from Acquired Immunodeficiency Syndrome (AIDS) that shows a significant rising tendency. The fight against malaria remains one of the biggest challenges of public health (Okara and Khalic, 1993). The ABO blood groups are not linked to the incidence of simple malaria infection but have been associated with rosette formation (Fischer, 1998). However, it has been widely reported that antigens present on erythrocytes in ABO are in volved in the susceptibility of red cells to species of plasmodium (Dacie and Lewis, 1991). Multiple human mutations associated with survival advantage in *P. falciparum* have been developed. These include structural haemoglobinopathies (HbS, HbC) quantitative haemoglobinopathies (the thalassemias) (Zaino, 1965), membrane mutations (spherocytosis, elliptoatosis, ovalocytosis)

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(Allen *et al*., 1999) and enzymopathies (Glucose-6-phosphate dehydrogenase deficiency) (Kay *et al*., 1992). Common haemoglobin-opaties in Africa are limiting factors to the severity of malaria infection by *P. falciparum* (Nnochiri, 1975).

Despite every effort to reduce or eradicate this disease it seems the achievement of the vision is far from being achieved. So in this community it is necessary to find out the distribution of malaria parasitemia and bacteramia among people that are showing symptoms of malaria considering all genders and age. Also, to determine if there is any association between malaria parasitemia, bacteramia, ABO blood group system and haemoglobin genotype as a measure to reducing the menace and also to assist policy makers to understand the need of the populace and plan accordingly.

MATERIALS AND METHODS

Study area: This study was carried out in three different hospitals namely: Obafemi Awolowo University Teaching Hospital, Seventh Day Adventist Hospital and Osun State Hospital Management Board all in Ile-Ife, Southwest Nigeria. The research was carried out between April and November, 2006. The subjects who were patients suspected for malaria infection in the said Hospitals which included both inpatient and out patient were recruited into the study.

Collection of sample: ABO ut 6 mL of venous blood was obtained from the antecubital vein by means of a sterile needle and syringe from each subject in which 5 mL was aseptically put into 20 mL glucose blood culture broth with the remaining 1 mL transferred into an EDTA container.

Sample processing: Thick and thin blood films were made and stained with Giemsa as described by WHO (1991). The slides were examined microscopically under oil immersion objective. For bacteramia, glucose broths into which blood was inoculated were incubated at 37°C for the period of 1-7 days aerobically and examined periodically (macroscopically) for gas production, turbidity and clot formation, which are indicators for growth. The broths were subcultured after 48h of incubation on MacConkey agar, Blood agar and Chocolate agar. The subcultured Blood agar and MacConkey agar plates were incubated aerobically at 37°C overnight while the Chocolate agar plates were also incubated overnight under 10% Carbondioxide (CO_2) condition using candle jar method according to the standard techniques (Cheesbrough, 2001). There was no facility for anaerobic isolation. Growth on culture plates were identified by colony morphology, gram stained and also characterized by standard biochemical test (Cheesbrough, 2001).

ABO Blood grouping and Haemoglobin electrophoresis were determined according to method described by Dacie and Lewis (1991).

RESULTS

Table 1 shows the distribution of malaria parasitemia and Bacteramia among various age groups. The result revealed that out of 515 cases investigated, 415 (80.6%) were positive for malaria parasitemia while 8 (1.6%) were positive for bacteramia. The age group 1-10 years had the highest prevalence of malaria parasitemia (91.7%) while the least was found in the age group 21-30 years (70.9%). Females (89.2%) were more infected than males (72.6%).

Distribution of malaria parasitemia by ABO blood group system and Haemoglobin genotype is shown in Table 2. Blood group O had the highest frequency (90.0%) while the lowest was in blood group AB (31.3%). Hb genotype AA had the highest prevalence rate of 85.2%, followed by AS (75.0%), then AC (69.2%) and the least was SS (44.0%). Table 3 shows the distribution pattern of Bacteria in which *Staphyloccus aureus* had the highest frequency (62.5%) while the *Klebsiella* sp. was the least

Table 1: Distribution of malaria parasitemia and bacteramia by sex and age

		Parasitemia		Bacteramia	
Parameter	Total no. examined	No. positive	(%) positive	No. positive	(%) positive
Age					
1-10	132	121	91.7	4	3
11-20	90	72	80	1	1.1
21-30	110	78	70.9	1	0.9
31-40	82	60	73.2	0	0
>40	101	84	83.2	2	1
Total	515	415	80.6	8	1.6
Sex					
Males	266	193	72.6	4	1.5
Female	249	222	89.2	4	1.6
Total	515	415	80.6	8	1.6

Table 2: Distribution of malaria parasitemia by ABO blood group system and Hb genotype

Parameter	No. examined	No. positive	(%) positive
Blood group			
A	170	125	73.5
B	60	43	71.7
AB	16	5	31.3
O	269	242	90
Total	515	415	80.6
Hb genotype			
AA	365	311	85.2
AS	112	84	75
AC	13	9	69.2
SS	25	11	44
Total	515	415	80.6

Table 3: Distribution pattern of bacteria isolates

Bacteria	No. of isolates	(%) of isolate
Staphylococcus aureus	5	62.5
Escherichia coli	2	25.0
Klebsiella sp.	1	12.5
Total	8	100.0

Table 4: Density of parasitemia in different age group

Age group (Year)	+(%)	++(%)	+++(%)	++++(%)
1-10	32(15.3)	36(38.7)	16(32.0)	37(57.8)
11-20	28(13.5)	12(12.9)	9(18.0)	23(35.9)
21-30	36(17.3)	24(25.8)	15(30.0)	3(4.7)
31-40	46(22.2)	9(9.7)	4(8.0)	1(1.6)
>40	66 (31.7)	12(12.9)	6(12.0)	0(0.0)
Total	280(100)	93(100)	50(100)	64(100)

Key: +-1-10 parasites/100 High Power Field (HPF), ++ -11-100 Parasites/ 10 (HPF), +++ -1-10 parasites every power field, ++++ - >10 parasites in every high power field

Table 5: Density of parasitemia in relation to Hb genotype

Hb Genotype	+(%)	++ (%)	+++ (%)	++++ (%)
AA	112 (53.9)	66 (70.9)	44 (90.0)	60 (95.3)
AS	60 (28.9)	22 (23.7)	4 (8.0)	3 (4.7)
AC	17 (8.2)	3(3.2)	1 (2.0)	0 (0.0)
SS	19 (9.0)	2 (2.1)	0 (0.0)	0 (0.0)
Total	208 (100)	93 (100)	50 (100)	64 (100)

(12.5%). Density of parasitemia in different Age group is shown in Table 4. The highest malaria density was obtained in the age group 1-10years. Density of parasitemia decreases as the age group increases. AA genotype had the highest density while both AC and AS were the least density (Table 5).

DISCUSSION

Malaria has continued to remain a major disease in tropical homogenous black African population. This infection is associated with great morbidity and mortality since the discovery of this infection many decades ago; it is still a great threat to both tropical and sub-tropical Africa. In this study, 415 (80.6%) out of 515 samples were positive for malaria parasitemia. This was in agreement with report of Alaribe *et al.* (1998) and Adefioye *et al.* (2007) with prevalence rate of 78.8 and 85%, respectively.

The high prevalence of malaria parasitemia is alarming and worrisome and may be due to many factors such as warm weather which favours the development of the insect vector, myriad of pools water in pot holes, abandoned tin and containers which are good breeding sites for the female anopheles mosquito, the insect vector (Guggemoos *et al.*, 1981). Another important factor may be because many people are asymptomatic and serves as temporary reservoir to mosquito therefore, distributes the parasite freely. Large percentage of the population and poor and cannot afford healthy diet to build strong immunity for resistance against malaria. Overcrowding also assists the fast spread of the parasite since a single infected mosquito can bite and inject sporozoites into the blood of a whole family within a very short period. Social and religions activities afford constant contact with the insect vector may contribute to high prevalence of parasitemia; this is witnessed in people attending night vigil and social gathering in the night in open places thereby exposing them to insect bite. Also occupational activities such as forming, fishing and night security services bring many people in direct contact with the insect vector as a result of constant exposure.

Those in age group 1-10 years were mostly affected with prevalence rate of 91.7% while age group 21-30 years was least infected. The age group 1-10 years with this rate conforms to pattern found in most endemic area and this reflects the low immunity in the group (Agbolahor *et al.*, 1993). The age group 21-30 years had 70.9% which is the least, which may be due to financial independence to feed well and education to administer self medication without visiting hospital. They visit only when the parasite is resistant to treatment. Furthermore, their level of immunity is high. Although, this variation is not significant when compare with other age groups aside from age 1-10years which is the highest.

The higher prevalence of 89.2% in female observed in this study is not significantly different ($p>0.05$) from male with 72.6% but the reason may be due partly to increased number of individuals that visit hospital during the study. This report contradicts the result of Agbolahor *et al.* (1993) who reported higher prevalence of 56.8% is male. This variation may be due to the subject and geographical location because he worked on the University Campus students. The higher rate in female may be physiological especially in adult female during menstrual period in cases of prolonged bleeding or multiple menstrual cycles, apart from blood loss, the stress, pain can lead to depression in immunity which may result into relapse or making them more susceptible (Wood, 1975).

This study has shown that (90.0%) prevalence rate was obtained among blood group O while the least was obtained in blood group AB. The high prevalence in blood group O observed is supported by Christina and Walter (2007). This is because during infection with *P. falciparum,* blood group O offers a survival advantage; group A confers a disadvantage and group B and intermediate effect. The ratio of group O to A is therefore, in geographic region where malaria is currently or was previously endemic. High prevalence of group O coupled with a low prevalence of group A is found throughout sub-Saharan Africa, where *P. falciparum* persists. Also in the Western hemisphere; the distribution of group A and group O generally matches malarials tropical distribution.

This finding is also corroborated by the fact that parasitized Red blood cells (Rbcs) form rosettes more readily with Rbcs belonging to group A or B than with Rbcs belonging to group O even in fresh clinical isolates (Rachanee *et al*., 1993).

Result obtained in this study revealed the prevalence rate of 85.2, 75.0, 69.2 and 44.0% belonged to genotype AA, AS, AC and SS, respectively. The higher prevalence of 85.2% in genotype AA is consistent with the earlier work of Emeribe and Osun (1993) who reported that, subject with AA had the highest incidence of malaria parasitemia. There are many hypothesis for the higher prevalence rate among genotype AA, for instance, Pasvol (1980) stated that the development of malaria parasite in blood requires adequate oxygen supply which is abundant in genotype AA against the low oxygen tension obtained in SS or AS genotype hence, higher prevalence is expected in AA homozygous. No confirmed evidence on the mechanisms by which the haemoglobin genotype AS protect against severe malaria has yet been documented (Rachanee *et al*., 1993).

Malaria parasite and bacteria co-infection revealed that it is a very rare case i.e., it is not a usual occurrence (Emeribe and Osun, 1993). The low prevalence of 1.6% bacteramia obtained in patient with malaria parasitemia may be considered to be a secondary infection as a result of severe and prolonged malaria infection.

The malaria parasite could be controlled by massive education at all level by the government such as home, school and worship centres. Every effort must be made to prevent human and vector contact, particularly the people in high risk group e.g., the infants, young ones, people with genotype AA and individual with blood group O. This should include the use of chemical impregnated nets, window nets etc. Breeding sites should also be destroyed while all stages of mosquitoes must be attacked by all means. It is also advisable for people to go to for medical diagnosis before it becomes severe; this will assist in controlling drug resistant strain. In all, anti malaria drugs should be administered once it has been confirmed.

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