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PLASMODIUM FALCIPARIUM PARASITEMIA IN PREGNANCY IN RELATION TO MATERNAL ANAEMIA

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ABSTRACT

This study was aimed at examining existing relationship between peripheral parasitaemia of *Plasmodium falciparum* and anemia among pregnant women in a secondary hospital and a tertiary hospital in Osogbo, South-Western, Nigeria. Two hundred and twenty five (225) patients were enrolled into this study, one hundred and fifty (150) from Asubiaro General Hospital, Osogbo and seventy five (75) from LAUTECH Teaching Hospital, Osogbo. A total of 30 (13.3%) women carrying the first pregnancy (primigravida), and 195 (86.6%) multiparous women (2-5) were enrolled. Mean age of recruited women was 31.511± SD 1.03, mean gestational age was 2.4267 ± SD 0.72 and mean packed cell volume was also 26.889 ± SD 0.43. Overall prevalence of malaria parasitemia was 63.6% while mean malaria parasite density was 461.33 among women infected with malaria parasite. Prevalence of malaria in pregnancy was highest amongst women with first pregnancy and in the age bracket 26 – 30 years (26.7%) and least among women greater than 40 years. Parasitemia decreased as parity increased, as women acquire immunity to malaria progressively with multiple pregnancies. Mild to moderate anaemia was also found to be prevalent among primigravida (11.6%) and this was associated with malaria parasitemia among these women .No correlated relationship was established between malaria parasitemia and age, gravidity, trimester of pregnancy, and Packed cell volume. Malaria chemoprophylaxis and other methods of malaria control should be sustained and advocacy for inclusion of malaria treatment in safe motherhood should be continued because of its beneficial potentials.

Key words: Malaria, Pregnancy, anaemia.

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INTRODUCTION

Malaria has been described as a disease of poverty and underdevelopment (1). It remains a complex and overwhelming health problem, with 300 to 500 million cases and 2 to 3 million deaths per year (1). About 90 percent of all deaths attributable to malaria

occur in sub-Saharan Africa. Although 40 percent of the world's population is at risk for malaria, in pregnant women the disease

has been most widely evaluated in sub-Saharan Africa.

Plasmodium falciparum infection during pregnancy increases the chance of maternal anemia, abortion, stillbirth, prematurity, intrauterine growth retardation, and low birth weight (defined as a weight of 2500 g), the greatest risk factor for neonatal mortality (2). Malaria during pregnancy is therefore a serious problem in sub-Saharan Africa, affecting an estimated 24 million pregnant women annually (2). Although *P. falciparum* infection in pregnancy could be asymptomatic, it often contributes to adverse perinatal outcomes with a high risk for infant death, maternal morbidity, including fever and severe anemia, abortion, and placental malaria particularly in areas of lower malaria endemicity (3). On the other hand anaemia in pregnancy is thought to be one of the commonest problems affecting pregnant women in developing countries.

In 1993, the World Bank ranked anaemia as the 8th leading cause of disease in girls and women in the developing world (4). Data collected from all over the world indicate that a total of 2170 million

people (men, women and children) are anaemic by WHO criteria (4). The most affected groups, in approximately descending order are pregnant women, the elderly, school children and adult men. In developing countries, prevalence rates in pregnant women are commonly estimated to be in the range of 40-60%. Among non-pregnant women this is 20%-40% and in school aged children and adult men the estimate is around 20% (2).

In sub-Saharan Africa, it is estimated that between 200,000 and 500,000 pregnant women develop severe anemia as a result of malaria (3). *P. falciparum* malaria in pregnancy is the primary cause of up to 10,000 maternal anemia-related deaths in sub-Saharan Africa annually (4). However, there have been conflicting reports from parts of sub-Saharan Africa on the relationship between placental malaria and maternal anemia. An earlier report from the Ubangi district of Zaire noted that malarious placentas had no consistent relationship to maternal anemia (5).

In other studies, maternal anemia and placental malaria were associated in all gravidity and age groups, with maternal anemia higher among women with placental malaria than those without placental malaria (3). This study was therefore aimed at examining existing relationship between peripheral parasitaemia and anemia among pregnant women in a secondary and one tertiary institution in Osogbo, South-Western, Nigeria.

MATERIALS AND METHODS

STUDY LOCATION

The study was carried out in Olorunda Local Government with headquarters in Osogbo, the capital of Osun State, Nigeria. Osogbo is in the tropical rain forest belt of Southwestern part of Nigeria, it is about 500kilometers from Abuja the capital city of Nigeria. It lies approximately on latitude 40^0N of equator and longitutude 7.34^0E of Greenwich meridian. The sites selected for this study are Ladoke Akintola University of Technology Teaching Hospital, and General Hospital, Asubiaro, in Osogbo.

STUDY SUBJECTS

The study subjects consisted of 250 pregnant women on regular ante-natal visit at Ladoke Akintola University of Technology Teaching Hospital, and General Hospital, Asubiaro, Osogbo between December 2008 and June, 2009. The women were of varying age ranging from 18-45 years.

BLOOD SAMPLE COLLECTION

Safety procedures were adopted in the collection of venous blood samples by swabbing the ante cubital fossae with 70% alcohol and 5mls of blood was drawn into EDTA bottle with sterile hypodermic needle. Thick and thin films were made on clean slides and labelled accordingly as recommended by WHO (3).

MICROCOPIC EXAMINATION

The thin films were fixed with methanol and all films were stained with 3% Giemsa stain of pH 7.0 for 30 min as recommended by WHO (6). Taking the number of leucocytes per micro liter of blood as 8,000, parasite density of blood using the thick film was expressed as: parasite count (x) 8,000 divided by number of WBCs counted. The thick films were used to determine the parasite densities while thin films were used to identify the parasite species and infective stages. Stained slides were examined under the light microscope using x100 objective lens (immersion oil) (3).

HAEMOGLOBIN DETERMINATION

Five ml of blood were collected inside EDTA bottle .Non heparinised capillary tubes were filled with blood sample from the EDTA bottle.The tip of the capillary tubes were cleaned with cotton wool and they were arranged inside the haematocrit centrifuge.They were centrifuged at revolution per min. The Packed cell volume was determined by using haematocrit reader to read the level of the haemoglobin (7).

STATISTICAL ANALYSIS

Data was analyzed using Statistical programmed for service solution (SPSS) 16.0 (SPSS Chicago Inc., IL, U.S.A.), the statistical significance of variables was estimated using chi-square test. Pearson correlation analysis shall be used to establish possible relationship or correlation between *Plasmodium falciparum* parasitemia and gravidity, age, trimester, packed cell volume.

P-values of equal to or less than 0.05 will be taken as measures of significance.

RESULTS

Two hundred and seventy five (275) women were enrolled into this study, one hundred and fifty pregnant women (150) from Asubiaro General Hospital, Osogbo and seventy five pregnant women (75) from LAUTECH Teaching Hospital, Osogbo. A total of fifty (50) ages matched, non-pregnant women were recruited as controls. They had no symptoms of malaria, and were recruited among members of staff, students and traders within the Teaching Hospital complex.

A total of 30 (13.3%) women carrying the first pregnancy (primigravida), and 195 (86.6%) women with multiple parity (2-5) (multigravida) were enrolled. All enrolled women in this study are married. Mean age of recruited pregnant

TABLE I: Prevalence of Malaria according to age

	Pregnant women n=225		**Non pregnant women n=100**	
Age (year)	Frequency (%)	Positive (%)	Frequency (%)	Positive (%)
16-20	11(4.9)	7(3.1)	3(3.0)	0(0)
21-25	44(19.6)	33(14.7)	14(14.0)	1(1.0)
26-30	95(4.22)	60(26.7)	22(22.0)	5(5.0)
31-35	52(23.1)	30(13.3)	20(20.0)	3(3.0)
36-40	21(9.3)	11(4.9)	30(30.0)	20(20.0)
41-45	2(0.9)	2(0.9)	11(11.0)	5(5.0)
Total	**225(100)**	**225(100)**	**100(100)**	**34(34)**

women was 31.511± SD 1.03 (controls; 32.22± 1.32) mean gestational age was 2.4267 ± SD 0.72 and mean packed cell volume was also 26.889 ± 0.43 for patients while controls was 35.5± .27405 .

Overall prevalence of malaria parasitemia was 63.5% for pregnant women while prevalence among controls was (12.0%). Meanwhile, means of malaria parasite density accounted for 461.33 among pregnant women infested with malaria 100 (100) 31(31) parasite in this study, same was 22.40 among non-pregnant control women.

Table I shows age distribution of recruited pregnant women in relation to malaria positivity. Women between ages 26 – 30yrs accounted for 42.2% of the study population. Followed by 31 – 35yrs which accounted for 23.1% .Other age distributions are 16 – 20yrs – 4.9%, 21 – 25yrs – 19.6 %, 36 – 40 - 9.3 % and > 40yrs - 0.9% . Prevalence of malaria in pregnancy was highest amongst women in the age bracket 26 – 30 years (26.7%), same was 2.0% among non pregnant controls. Prevalence was least among women greater than 40 years. Statistically however, there was no significant difference in the trend. ($\chi 2$ = 5.54, df =5, p >0.05).

Table II: Prevalence of malaria according to Gestational age

Gestational Age	**Frequency (%)**	**No. +ve for malaria (%)**	**No –ve for malaria (%)**
1st Trimester	**31 (13.8)**	**20 (8.9)**	**11 (4.9)**
2nd Trimester	**67 (29.8)**	**41 (18.2)**	**26 (11.6)**
3rd Trimester	**127 (56.4)**	**82 (36.4)**	**45 (20)**
Total	**225 (100.0)**	**143 (63.6)**	**82 (36.4)**
X^2 = 0.23, df =2, p>0.05			

Table III: Prevalence of Malaria according to Parity

Parity	**No. Examined**	**No. +ve for Malaria**	**No. -ve for Malaria**
1 + 0	**92**	**58(25.8)**	**34 (15.1)**
2+ 0	**68**	**45 (20)**	**23 (10.2)**
3 + 0	**39**	**25 (11.1)**	**14 (6.2)**
4 + 0	**18**	**11(4.9)**	**7(3.1)**
≥ 5	**8**	**4 (1.7)**	**4 (1.7)**
Total	**225**	**143 (63.6)**	**82 (36.4)**

Table IV: Relationship between PCV and malaria

PCV(%)	No. Examined (%)	Patients		Controls	
		No. +ve for malaria(%)	No. - ve for malaria (%)	No. + ve for malaria (%)	No. -ve for malaria (%)
10-20	2 (0.9)	0 (0)	2 (0.9)	0 (0)	0 (0)
21-30	66 (29.3)	47 (20.9)	19 (8.4)	3 (6.0)	5 (10.0)
31-40	157 (69.8)	96 (42.7)	61 (27.1)	3 (6.0)	39 (78.0)
Total	225 (100.0)	143 (63.6)	82 (36.4)	6 (12.0)	44 (88.0)

Table II is a frequency table which shows the relationship between gestational age (trimesters) and prevalence of malaria positivity. One hundred and twenty seven women (56.4%) were in the third trimester, while 29.8% (67) were in the second trimester. Meanwhile only 13.3% (30) were in their first trimester. Prevalence of malaria positivity increased progressively across trimesters, from first (8.9%), second (18.2%) to third trimester (63.5%).However, there was no existing significant difference between malaria positive and negative cases with respect to trimester. ($\chi 2 = 0.23$, df =2, $p > 0.05$).

Malaria parasitemia was prevalent among the primigravida (women with first pregnancy) 58/225 (25.8%) and parasitemia decrease as parity increases, among the secondigravida (women with second pregnancy) prevalence was found to be 20% (45/225) while Parity 3, 4, and 5 have a prevalence of 11.1% (25/225) ,4.9% (10/225) and 1.7% (4/225) respectively as shown in table III.There was no significant difference between malaria positive and negative cases with respect to parity . ($\chi 2 = 0.89$, df =4, $p > 0.05$).

Mild to moderate anemia (PCV; 21 - 30) as found in the study population accounted for 29.3% (66/225) and was prevalent, while severe anaemia was uncommon 0.9% (2/225) in the study population. A large population of pregnant women had PCV 31 – 40% as shown in the frequency table IV. Of the 2 patients that had severe anaemia (PCV≤ 20), none was positive for malaria parasite, While 44 (19.6%) of 65 patients that had moderate anaemia demonstrated malaria parasite positivity. Pearson Chi-square test showed no correlation between malaria positive and negative cases in relation to packed cell volume. (($\chi 2 = 5.55$, df = 2, $p > 0.05$). (Table IV).

DISCUSSION

Malaria during pregnancy is a serious problem in sub-Saharan Africa, affecting an estimated 24 million pregnant women. Pregnancies in women living in malaria endemic regions, particularly in sub-Saharan Africa are associated with a high frequency and density of *Plasmodium falciparum* parasitaemia, with high rates of maternal morbidity including fever and severe anaemia, with abortion and stillbirth, and with high rates of placental malaria and consequently low birth weight in newborns caused by both prematurity and intrauterine growth retardation (8).

In highly endemic malarious area where semi-immune adults usually have substantially acquired resistance to local strains of plasmodia, the prevalence of clinical malaria is higher and its severity greater in pregnant women than non-pregnant women (2). This was found true in this study, as prevalence of malaria parasitemia was found to be 63.6% which was higher than prevalence of 12.0% among non pregnant women used as control. This prevalence is similar to 72% found by Adefioye *et al* (9) in a study among pregnant women in Osogbo. It is also worth mentioning that Okwa in Lagos (10) found a comparable prevalence of 60%. However, Huddle *et al* (11) in rural Malawi study found extremely high prevalence of 83%. In studies conducted by Brabin in 1991 (12), the primigravidae were more susceptible to malaria infection than the multigravidae, which was also confirmed by this study. Prevalence was highest among the primigravidae (25.8%) and malaria positivity decreases as parity increased. Among the secondigravida, prevalence was found to be 20% while Para 3, 4, and 5 had a prevalence of 11.1%, 4.4% and 1.7% respectively. Onwere *et al*. (13) in

Aba found higher prevalence among primigravidae (39.3%).

Younger women appeared to be susceptible to malaria in this study as prevalence was highest among age group 26 – 30 (26.7%), a lower prevalence of 2.0% was found among the non pregnant controls. This contradicts the findings of Adefioye *et al* (9) that found 36 – 39 year age group to be more susceptible. However 3rd trimester prevalence in this study is a deviation from previous studies as Brabin in 1983 (2) found in Western Kenya that prevalence was highest at 13-16 weeks gestation (1st trimester), and found similar number of recoveries in both groups during the 2nd and 3rd trimesters. The loss of immunity in early pregnancy was equivalent to an 11-fold decrease in the rate of recovery from infection. The recovery seen in late pregnancy suggests that the women mount a satisfactory immune response to malaria infection, reacquiring their pre-pregnancy immune status at about the time of delivery (14). However, reasons that may be adduced to high prevalence of parasitemia include the fact that, large numbers of women in this study were in their 3rd trimester, and registered late for care, which is extremely common in this environment.

It is not clear whether a natural immune depression in pregnancy or factors associated with the placenta encourage parasite multiplication and determines clinical manifestations. Pregnant women with malaria in pregnancy in Tanzania had significantly higher total and free serum cortisol concentrations than controls without malaria whether nulliparous or multiparous. High plasma corticosteroid levels may have an immunosuppressive effect on cell mediated immune responses. Cell-mediated immune responses to malaria antigens are more markedly suppressed in first than in subsequent pregnancies (2).

The multigravidae are presumably less affected because immunological memory from first pregnancy is retained. In first and second pregnancies women are especially vulnerable. Mcgregor (15) identified the factors responsible for susceptibility of primigravidae to malaria as inhibition of type 1 cytokine responses (interferon, interleukins 2 and 12 and TNF). At pregnancy, immunity has been altered; hence, with malaria 70–80% of pregnant women in malarious areas are susceptible to anaemia (2).

Mild to moderate anaemia (mean PCV: 26.1%, Control: 35.5%) was prevalent generally in this study (28.9%) and was highest in the 2nd trimmester. This prevalence seems low when compared with other studies. Reasons for this might be that most of these women are on routine folic acid, iron and other haematinics which might had brought up their PCV. Idowu *et al* (16) observed higher prevalence of anaemia in pregnanacy (76.5%) in Abeokuta because women attending traditional birth home (TBH) were recruited into his study. Most of the women were taking herbal remedies made from tree barks, leaves and roots of undisclosed plants.

The iron supplementation is necessary to prevent anaemic condition during increased physiological burden of pregnancy (2), same was lacking among women in the TBH, this might had contributed to the higher prevalence recorded in that study. The peak of anaemia recorded in this study (2nd trimester) also coincides with the period when haemodilution get to its maximum peak. This may have contributed to the high prevalence recorded in the 2nd trimester, showing that anaemia is further aggravated by haemodilution in addition to other possible factors. However, this finding contradicts the report of WHO in which anaemia is said to be significantly higher in the 3rd trimester of pregnancy than the first two trimesters. More so most of these women are in the 3rd trimester in which tendencies to anaemia is much reduces. This aggregation in 3rd trimester might be due to late antenatal booking.

CONCLUSION

This study recorded high prevalence of malaria parasitemia among pregnant women attending antenatal clinic in Osogbo. The study also recorded mild to moderate anaemia among pregnant women with malaria parasitemia compared to those without malaria parasite and the controls. The high prevalence of malaria parasites can be traced to the fact that Osogbo is in the rain forest belt of southwestern Nigeria, which is a good environment for mosquito breeding especially during the rainy season when this study was conducted.

Malaria parasitemia and its attendant's complications have been demonstrated to be worst in pregnancy in this study. Proper malaria control programme is highly necessitated among this highly susceptible group especially in the tropics. With good ambient temperature, humidity, and mosquito will conveniently transmit malaria. There are stagnant pools and blocked drainages in Osogbo, many of the pregnant women came from shanty region of the city. Pregnant women must therefore be

adequately protected from double edged sword of malaria and anaemia. Advocacy for inclusion of malaria treatment into Safe Motherhood Initiatives must be sustained, because of these overwhelming implications of malaria parasitemia in pregnanacy.

It is recommended that routine intermittent preventive treatment of malaria is therefore recommended in this environment, for women in pregnancy. Several studies have shown that protection against malaria contributes to the prevention of anaemia in pregnancy, thus highlighting the importance of chemoprophylaxis and use of other methods of malaria control like insecticide impregnated bed net. These good practices must however be sustained and recommended to be included in Safe Motherhood Initiatives because of its beneficial potentials in our resource poor setting. Early ante natal booking for effective monitoring and prompt treatment of both malaria and anaemia in pregnanacy will contribute significantly in reducing maternal mortality and morbidity and its attendant perinatal mortality. Regular environmental sanitation to dislodge mosquitoes from their breeding places will also go a long way to reduce prevalence of malaria in shanty towns and villages commonly seen in the tropics which we belong.

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