Original Research Article

IJCRR
Section: Healthcare Sci. Journal
Impact Factor 4.016
ICV: 71.54

Prevalence of asymptomatic malaria parasitaemia during pregnancy and its effect on foetal birth weight

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ABSTRACT

Background: Pregnant women in endemic area may experience malaria infection without clinical symptoms. Its effects on the neonatal outcomes mayalso occur in this asymptomatic state.

Objective: To determine the prevalence of asymptomatic malaria parasitaemia in pregnancy and the relationship between the level of malaria parasitaemia and foetal birth weight.

Materials And Methods: A total of 290 asymptomatic paturients and their babies were recruited over 4 months with informed consent. The maternal, placenta and cord blood samples were obtained and examined for level of malaria parasitaemia. Newborns were weighed and classified as normal birth weight (≥2500 g) or LBW (<2500 g).Pearson correlation was used to determine the relationship between the levels of malaria parasitaemia and birth weights. Student's t and Pearson chi-square tests were used to compare means and percentages.

Results: The prevalence of malaria parasitaemia was 32.8%, 31% and 24.1% in the maternal, placental and cord blood smear respectively. The prevalence of low birth weight was 12.1% with women with malaria delivered more LBW babies (31.6%) than their uninfected counterparts (2.6%) (p=0.006). However, correlation showed a weak positive correlation between the levels of maternal parasitaemia(r = 0.163; p=0.504) and cord blood parasitaemia(r = 0.244; p = 0.400) but weak negative correlation with the level of placental parasitaemia (r = 0.135; p= 0.598) and foetal birth weight which were not statistically significant.

Conclusion: There was no significant correlation between the level of malaria parasitaemia and birth weight in asymptomatic parturients. However, the main impact on pregnancy outcome was the higher prevalence of LBW.

Key Words: Malaria parasitaemia, Pregnancy, Foetal birth weight

INTRODUCTION

Malaria remains a major health concern worldwide; perhaps the most important parasitic infection affecting mankind with an estimated 3.3 billion people at risk of malaria in 2010 worldwide.[1] An estimated 655,000 deaths were recorded globally in 2010 of which 86% were children less than 5 years of age. The disparity in region specific mortality is huge with 91% of all deaths recorded by WHO in Africa region.[1] Women are more susceptible to malaria during pregnancy and in the puerperium.[2, 3]

In areas where malaria is endemic, that is stable malaria transmission like Nigeria, at least one in four pregnant women has evidence of peripheral or placental malaria at delivery.[4, 5] However, most cases of malaria in pregnancy in such areas may remain asymptomatic thus undetected and untreated.[6] The pre-existing immunity retained during previous exposures protect against clinical malaria. Unfortunately, this subclinical infection poses great challenge to mother and foetus.[4, 7]

The mechanism underlying increased susceptibility to malaria and the severity of the disease in pregnancy is not fully

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Received: 03.08.2016 **Revised:** 04.09.2016 **Accepted:** 10.10.2016

understood.[8,9] It has been suggested that despite the acquired antimalaria immunity of these pregnant women, the uteroplacental vascular space apparently provides a site for parasite sequestration and development.[10,11]

The main effects of malaria in pregnancy on birth outcomes are thought to be mediated by maternal anaemia[12,13,14] and placental insufficiency.[11,15] Both factors have been suggested to act together to cause either intrauterine growth restriction (IUGR) or preterm delivery leading to low birth weight (<2,500g).[16,17,18] Malaria is one of the causes of severe anemia in pregnancy.[19] Over 26% of anaemia in pregnancy is attributed to malaria, and malaria related maternal deaths are reaching an unacceptable rate of 23%.[5,20]

Low birth weight (<2,500g) is known to be an important risk factor for infant mortality, an important cause of foetal and neonatal morbidity and one important determinant of infant healthy growth and development.[4,13,21] Up to 20% of LBW in sub-Saharan Africa has been attributed to malaria in pregnancy and this figure represents 35% of preventable low birth weight in the region.[8, 12]

The prevalence of asymptomatic malaria parasitaenia in pregnancy and its effect varies even within the endemic region as reported by various workers.[21,22,23] In Nigeria, few studies focusing on malaria in the peripartum period have been conducted but with variable findings.[23,24,25]

Malaria in pregnancy still remains a major health concern in the Sub-Saharan Africa. Malaria is thought to be an important contributor to the 3.5 million LBW babies born annually in sub-saharan Africa,[8] attributable fraction is estimated to be 19%.[8,23] Most of the studies done focusing on peripartum malaria parasitaemia in Nigeria did not distinguish the effect of an asymptomatic parturient, thus the importance of asymptomatic carrier is poorly elucidated. Therefore, this present study was designed to study prevalence of asymptomatic peripartum malaria parasitaemia and its effects on birth weight.

MATERIALS AND METHOD

The study took place at the Federal Medical Centre, Owo is a tertiary health centre, located Owo, Ondo State, South West Nigeria. Ondo State is located entirely within the tropics with annual rainfall between 150mm and 2,000mm.[27,28] Malaria transmission in Southwest Nigeria is perennial but seasonal and peaks during the rainy season, which normally runs from April to November. The study received ethical approval from Federal Medical Centre, Hospitals Management Board ethics review committees.

The study was a prospective study using a purposive non-probability random sampling method, there was recruitment of all consenting parturient in labour that satisfied the inclusion criteria until the desired sample size is completed. Their babies were weighed at birth, samples obtained and the proforma filled. Inclusion criteria were confirmation of active phase of labour; asymptomatic, non-febrile parturient and singleton pregnancy without any known congenital anomaly. Exclusion from the study are subjects that refused to participate in the study; those on anti-malaria at the time of labour; presence of any medical illness such as diabetes mellitus, chronic renal disease, haemoglobinopathies, HIV/AIDs, chronic hypertension and severe pre-eclampsia/ eclampsia and presence of multiple gestation, intrauterine foetal death and obvious foetal anomalies. There were 290 women recruited having met the inclusion criteria.

Sample collection

In labour, 5mls of blood sample was collected from the mother, 3mls of blood was collected from the clamped cord immediately after delivery and placenta blood was also obtained into EDTA bottles. The placental weight, birth weight and pregnancy outcome was recorded. In this study, malaria infection was taken as the presence of asexual *P. falciparum* parasites of any density, in a thick film.[29,30]

Analytical methods

Hemoglobin (Hb) was measured using the method of Schoen and Solomon[17] and determination of packed cell volume (PCV) by the microhematocrit method. Malaria parasitaemia was estimated in thick and thin film from maternal blood, cord blood and placental aspirate of all samples collected, according to the method described by Cheesbrough.[31] Peripheral blood, parasite density was determined by counting asexual forms of the parasite per 100 white blood cells (WBC) converted to parasites/µL using a predetermined blood sample total white blood cell count.[31,32] Parasite density was graded as Low (parasites < 1,000/µL), Moderate (1,000- 4,999/µL) and high (>5,000/µL).[33] Quality Assurance was maintained by randomly selecting 5 slides in a pool of every 30 slides for comparison in the institution's main laboratory. The babies were weighed within 30 minutes of birth after drying the body using a bassinet weighing scale (Salter model 180 made in England 2002-0218015B) with maximum 15kg capacity and scale precision of 50g

Statistical Analysis

Data was analysed using the (SPSS) Version 17 (SPSS Inc, Chicago, IL). Analysis included the use of descriptive statistics such as means and standard deviation of age, parity and haemoglobin concentration. Using student t-test and chi square test for association between maternal and babies' characteristic and malaria parasitaemia were determined. P-value of ≤ 0.05 was taken as significant. Multivariate analysis using linear regression of maternal age, parity, previous antimalaria use and maternal parasitaemia was done. Expres-

sion of percentages, frequency tables and charts was used to illustrate the presence of parasitaemia (maternal, cord and placenta) and risk factors for LBW. Pearson correlation was used to determine the relationship between the levels of parasitaemia and birth weights, these were shown on a scatter plots.

RESULTS

Sociodemographic background of the study population is shown in Table 1; with 290 women who met the inclusion criteria were recruited for the study. Their age ranged from 18- 44 years with mean age of 30.69±5.52 and modal age of 33. The mean packed cell volume (PCV) at delivery was 32.38±3.145% and ranged from 26 to 40%. All babies were delivered alive with no still birth in this study. Mode of delivery; 70.7% of the labour had spontaneous vaginal deliveries with 29.3% ended up in a caesarean delivery as shown in Table 2. There were 95 women (32.8%) with positive maternal parasitaemia, 90 positive placental smears (31.0%) while positive cord blood parasitaemia were 70 (24.1%) as shown in Table 3. The cumulative prevalence of malaria infection at delivery (total number of women positive for malaria either by peripheral, cord blood or placental blood smear examination) was 29.3% (255/870). When the positive maternal parasitaemia was related to parity, the number of positive smears in nulliparous (30) 46.2%, primiparous (10) 18.2% and multiparous (55) 32.4% however, the differences were not statistically significant (P =0.346). The level of malaria parasitaemia is higher in the maternal peripheral blood smear compared with placental and cord blood smears as shown in table 3. The same difference was noted with the parasite density as shown in Table 4. The mean parasite density in maternal blood was 4,308±1896.67/µl, in placental smear was 1,853±1351.36/µl and in cord blood was 368±217.49/µl. Of the 290 cord samples collected, 70 (24.1%) were found to be positive for asexual forms of plasmodium falciparum, thus giving a prevalence of congenital malaria to be 24.1%. However, overall parasite density in cord blood is lower than both peripheral and placental parasitaemia density as show in Table 4.

Parasitaemia was associated with lower mean birth weight 3.03k g ±(0.615) versus 3.31k g ±(0.523) among mothers with peripheral parasitaemia; P = 0.257 as described in Table 5. Low birth weight (LBW) occurred in 35/290 deliveries thus giving a prevalence of 12.1%. Peripheral parasitaemia was also significantly associated with LBW babies (30/95 [31.6%] versus 97/195 [2.6%]; P =0 .006, OR = 1.6 [1.1–2.5]). Preterm deliveries contributed 14.3% of the LBW babies. Figure 1, 2 and 3 shows the correlation between the birth weight and maternal, placental and cord blood parasitaemia respectively. The correlation coefficient were; maternal=+0.163, p=0.504, placental= -0.135, p=0.594 and cord blood=+0.244, p=0.400. There was a positive correlation between parasite densities in the peripheral film and cord blood film. The correlation coefficient was +0.375, p=0.187. See Scatter plot illustration in figure 4.

DISCUSSION

Malaria in pregnancy contributes significantly to maternal and perinatal morbidity and mortality in sub Saharan African.[6, 8,26,34] This study illustrates the continuing impact of malaria in otherwise healthy asymptomatic pregnant Nigerian women, 32.8% of whom had peripartum malaria parasitaemia. This rate confirm recent reports in pregnant or at parturition in Nigerian women [6, 35, 36] although prevalence ranging from 12.5 to 80% [23, 35, 37] had been reported in Nigeria. The findings is also in agreement with findings from Cameroon, Senegal and Sierra Leone[38, 39, 40] but higher than in northern Nigeria.[26] Several factors could have accounted for the disparity in the prevalence like the season, study population characteristics (socio-economic class, parity, and age), the training of the microscopist, the use of chemoprophylaxis (IPT, ITN) and the study design. In this study, parasitaemia at the time of delivery was found to be associated with nulliparity and maternal age. Several other studies have reported similar associations.[6,41] The mean age of parasitized parturients in this study was significantly lower than the unparasitized parturients (29.79±7.43 was compared to 31±4.36) years (p = 0.001) as shown Table 5, similar observation was made by other investigators.[6,41] This suggested that pregnancy-associated immunity and naturally acquired immunity increases with age. The hypothesis was that the development of pregnancy associated immunity, for example, production of antibodies that inhibit adherence of placental parasites to chondroitin sulphate A, may be very important in women <25 years of age who have lower levels of acquired immunity. While older women living in such endemic areas due to repeated exposures, may have obtained adequate immunity and are thus less dependent on anticytoadherent antibodies. Parasitaemia in the mothers was found to be associated with lower maternal packed cell volume. This finding is not unusual as the association of malaria in pregnancy and low hematocrit has been recognized and reported by previous workers.[6] The drop in hematocrit occurs as a result of the fact that parasitized and unparasitized erythrocytes are destroyed by the spleen during malaria infection. It is however known that using of antimalaria drugs that are normally effective within a locality significantly reduces the occurrence of this anaemia.[42] Although the mean PCV of those parturient that had malaria parasitaemia was lower than those without parasitaemia (31.42% ±3.61 compared to 32.89%±2.85) the difference between the means was not statistically significant (p= 0.374) Table 5. This may be due to the fact that majority of the patients in this study used malaria chemoprophylaxis.

The main effect of maternal parasitaemia on the babies was the reduction in the birth weight. This is consistent with the observations from other malaria endemic countries.[6,38,43] The impact of malaria during pregnancy on LBW in sub-Sahara Africa has been extensively reviewed.[8] The LBW prevalence in this study was 12.1% and significantly increased with mothers with parasitaemia. This is within 8-25% range reported from previous studies from Sub-saharan Africa.[24, 44, 45]

Multivariate analysis showed that younger maternal age less than 20years, booking status, use of chemoprophylaxis and PCV were significantly associated with malaria parasitaemia in the women. Younger maternal age and hematocrit level were also described by other investiators.[6,26,39] These authors observed that younger but not older primigavidae were more likely to have placental malaria. This may explain the weak inverse correlation observed in this study with placental parastaemia and birth weight.

There was a positive correlation between parasite densities in the peripheral film and cord blood film. This implies that a population with increased prevalence asymptomatic maternal parasitaemia at delivery may have high prevalence of congenital malaria.

Chemoprophylaxis during pregnancy has been shown to reduce the risk of malaria infection in pregnant women significantly.[26] This was corroborated in this study with a significant association between chemoprophylaxis and reduction of malaria parasitaemia in cord blood (P= 0.05, RR= 0.0813) but reduction was not statistically significant for maternal and placental blood smears. This finding may be due to the fact that out of 91.4% (265 out of 290) that used chemoprophylaxis only 63.8% (185 out of 265) received the two doses of sulphadoxine pyrimethamine (SP) in contrast to high rate reported by some workers.[26] The two doses of IPT had been proven to be the currently most effective chemoprophylaxis for prevention of malaria during pregnancy in areas where transmission of Plasmodium falciparum malaria is stable like ours.[1, 26] The observed impact of malaria on the mother and their newborns add justification for promoting use of malaria preventive measure in pregnancy.

CONCLUSION

In Nigeria, one in every four asymptomatic women has malaria (maternal, placental and/ or cord) parasitaemia at delivery. Maternal age less than 20 years was the most important predisposing factor. There was no significant correlation between the level of malaria parasitaemia and birth weight in asymptomatic parturients. However, the main impact on pregnancy outcome was the higher prevalence of LBW.

ACKNOWLEDGEMENTS

Authors acknowledge the immense help received from the scholars whose articles are cited and included in references of this manuscript. The authors are also grateful to authors / editors / publishers of all those articles, journals and books from where the literature for this article has been reviewed and discussed.

Source of funding: None

Conflicts of interest: None

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Table 1: Characteristics of the study population

Characteristics	Frequency	Percent (%)
Age in years <20		
20 -35	15	5.2
>35	235	81.0
Mean ±SD 30.67±5.52(years)	40	13.8
Marital Status		
Married	270	93.1
Single	20	6.9
Parity		
Nulliparous	65	22.4
Primparous	55	19.0
Multiparous	170	58.6
Mean ±SD 1.91±1.525		
Previous Obstetrics History		
Not Significant	275	94.8
Previous LBW	10	3.4
Previous Preterm Birth	5	1.7
Chemoprophylaxis use		
IPT 1	80	27.6
IPT 2	185	63.8
None used	25	8.9
Use of ITN		
YES	35	12.1
NO	255	87.9

ITN=Insecticide Treated Net; IPT= Intermittent preventive treatment

Table 2: Labour History and Outcome

Characteristics	Frequency	Percent (%)
Onset of Labour		
Spontaneous	265	91.4
Induced	25	8.6
Mode of delivery		
SVD	205	70.7
C/S	85	29.3
PCV at delivery		
< 30%	35	12.1
≥ 30%	255	87.9
Mean PCV±SD 32.38±3.145		
Birth Weight		
LBW (<2.5kg)	35	12.1
NBW (2.5-4 kg)	235	81.0
Macrosomia (>4kg)	20	6.9
Mean birth weight ± SD 32.38 ± 3.145		
Mean Placental weight ± SD 0.589 ± 0.101		
SEX	145	50
Male	145	50
Female		

Table 3: Malaria parasitaemia in blood smears

Characteristics Malaria Parasite	Frequency	Percent (%)
In Maternal blood		
Present	95	32.8
Absent	195	67.2
In placental blood		
Present	90	31.0
Absent	200	69.0
In Cord blood		
Present	70	24.1
Absent	220	75.9

Table 4: Classification of malaria Parasitaemia density /µl

Malaria density/ µl	Maternal blood smear		Placental aspirate smear		Cord blood smear	
	Frequency	Percent (%)	Frequency	Percent (%)	Frequency	Percent (%)
No Parasites	195	67.2	200	69.0	220	75.9
< 1,000/ µl	-------	--------	30	10.3	70	24.1
1,000-1,500/ µl	10	3.4	15	5.2	------	------
>1,500 /µl	85	29.3	45	15.5	-------	-------

Table 5: Pregnancy Outcome in parasitized and non-parasitized

Characteristics	All Women	Parasitized	Non-parasitized	p-Value
Birth weight(Kg) mean±SD	3.21±0.559	3.03 ±0.615	3.31±0.523	0.259
PCV at delivery (%) mean±SD	32.38±3.145	31.42±3.610	32.89±2.845	0.374
Age (years) mean±SD	30.67±5.52	29.79±7.43	31.00±4.36	0.001*
Low Birth Weight	12.1%	31.6%	2.6%	0.006*

PCV= Packed cell volume, Significant *

Table 6: Maternal parasitaemia by Parity

Characteristics Parity	Maternal malaria Parasites		Total
	Present	Absent	
Nulliparous	30 (46.2%)	35(53.8%)	65
Primiparous	10 (18.2%)	45(81.8%)	55
Multiparous	55 (32.4%)	115(67.6%)	170
P Value= 0.342 RR(95% CI) = 0.322 (0.313-0.332)			

RR=Relative risk, CI= Confidence Interval

Table 7: Anaemia as a risk factor for Maternal Parasitaemia

Characteristics PCV (%)	MP (+ve)	MP(+ve)	Total	P value	RR
<30	30 (85.7%)	5(14.3%)	35(12.1%)	0.001*	
≥30	64 (23.5%)	191(74.5%)	255 (87.9%)		0.005

RR=Relative risk, Significant*

Table 8: Age as a risk factor for Maternal Parasitaemia

Characteristics Age (years)	MP (+ve)	MP(+ve)	Total	P value	RR
<20	14 (93.3%)	1(6.67%)	15(5.2%)	0.033*	
≥20	80 (29.1%)	195(70.9%)	275 (94.8%)		0.243

RR=Relative risk, Significant*

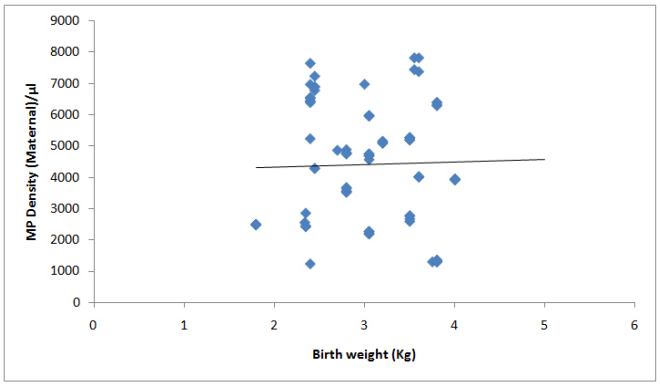

Figure 1: Scatter diagram of correlation between maternal malaria density and birth weight.
Correlation coefficient (r) = 0.163; p=0.504
MP= Malaria parasites

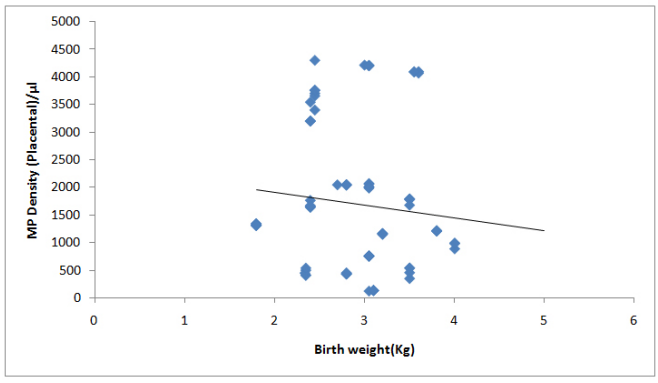

Figure 2: Scatter diagram of correlation between placental malaria density and birth weight.
Correlation coefficient (r)= - 0.135; p= 0.598
MP= Malaria parasites

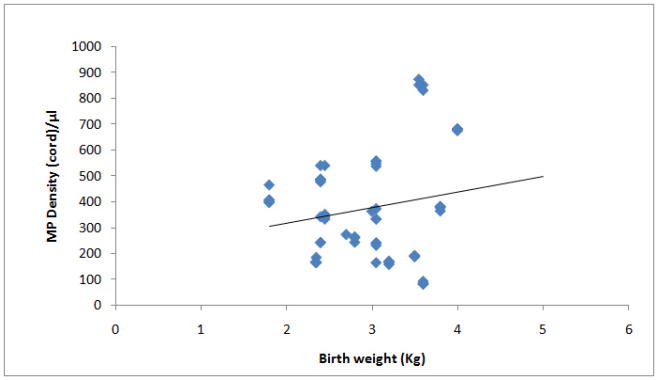

Figure 3: Scatter diagram of correlation between cord blood malaria density and birth weight.
Correlation coefficient (r)= 0.244; p = 0.400
MP= Malaria parasites

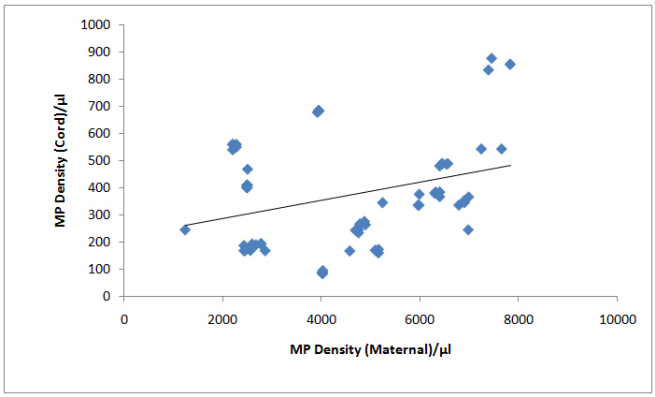

Figure 4: Scatter diagram of correlation between cord blood and maternal malaria density.
Correlation coefficient (r)= 0.375, p=0.187
MP= Malaria parasites