The Prevalence of Congenital Malaria in Secondary and Tertiary Health Care Facilities in Uyo, Nigeria

***Ikon GM, Akpabio RP, Ohagim PI, Etang UE, Udo EE, Moses AE**

ABSTRACT

A cross-sectional study of congenital malaria was carried out at two different hospitals within Uyo metropolis; St Luke's Hospital, Anua and University of Uyo teaching Hospital, Uyo. A total number of 218 blood samples were collected from the mothers and their new born babies and examined microscopically for malaria parasite using both thick and thin films. The prevalence rate of congenital malaria obtained in this study was 12.8%. The prevalence of placental, cord and maternal parasitaemia were 11.5% 12.4% and 3.7%, respectively. Maternal genotypes were determined and it was observed that mothers with haemoglobin AA showed a higher rate of parasitaemia than AS. Plasmodium falciparium was the only species encountered in this study. Malaria parasitaemia did not seem to have affected the faetal weight of the babies. A comparative analysis of the relationship between congenital malaria and maternal, cord blood and placental parasitaemia as obtained from this study, shows that placental parasitaemia had the highest prevalence (32.1%) of congenital malaria (p>0.05) in Uyo. The use of intermittent prevention therapy prompt management of cases and insecticide treated nets should be emphasized for all pregnant women.

Keywords: Malaria, Parasitaemia, Congenital, Plasmodium falciparium

INTRODUCTION

Congenital malaria can be defined as the malaria infection resulting from the transplacental transmission of malaria parasite in maternal blood (particularly, *Plasmodium falciparium*), to the new born during pregnancy or prenatally during labour[1]. In malaria endemic areas, including Nigeria, pregnant women are especially vulnerable to malaria. This is largely due to reduced cell mediated immunity that allows retention of foetal allograft which also interfers with resistance and enables the parasite to be transmitted to the foetus or to the babies during delivery through the placenta[2]. The placental malaria stimulates the production of inflammatory mediators resulting in a shift from Th2 cytokines immune responses which is usually associated with healthy pregnancy towards Th1 type cytokines immune responses[3]. The presence of ring form parasites in the erythrocytes of new-borns aged less than 7 days irrespective of clinical symptoms is evidence which determines the presence of congenital malaria[3,4].

Diagnosis of congenital malaria in the new born is usually carried out 7 days after birth or later if there is no incidence of mosquito bites or blood transfusion[3]. Microscopy is regarded as the gold standard method to diagnose malaria and determine which species of plasmodium causes the infection[4]. This is done by examination of a blood film under the microscope in a laboratory to reveal the distinctive physical characteristics of each species [4]. In *Plasmodium falciparum,* only early trophozoites or ring form parasites and gametocytes are seen in the peripheral blood. The presence of matured trophozoites or schizonts in peripheral blood smears is quite unusual as these are always confined to the tissues. However, symptoms usually occur 10-30 days post partum[5]. In about 80% of the cases of congenital malaria, the most common clinical features include anaemia, fever and spleenomegaly[6]. Other notable signs and symptoms of infection includes: heptomegaly, jaundice, regurgitation, loose stools and poor feeding. In rare cases, symptoms such as drowsiness, restlessness and cyanosis may also be seen[6].

There have been many studies on congenital malaria showing an increase in occurrence which has gone a long way in awakening public health concern globally[6,7]. Research has shown that increased burden of childhood malaria in endemic regions of the world is widely associated with malaria during pregnancy[7]. However, according to the World Health Report of 2015, about 214 million cases of malaria occurred worldwide[7]. In areas of high malaria endemicity, majority of the malaria cases are associated with morbidity and mortality in

Department of Medical Microbiology and Parasitology, Faculty of Clinical Sciences, University of Uyo, Uyo, Akwa Ibom State-Nigeria.

*Corresponding author:degraceikon@yahoo.com

young children below 5 years of age and this amounts to child mortality of nearly 1 million each year[8, 9, 10]. This means that a child dies of malaria every 20 seconds.

However, reports have shown that multigravid women seem to have resistance to *P. falciparum* than primigravid[9]. This comes as a result of acquired antibodies (Abs) against chrondroition sulphate (CSA), a novel receptor that does not commonly support binding of other parasites[7]. Due to acquired immunity, placental malaria is briefer in multigravid women and less inflammatory compared to that of the primigravid women. Reports have also shown that women with malaria infection of the placenta also have increased risk of passing HIV infection to their newborns[11].

MATERIALS AND METHOD

Study Population

The study population comprised mothers and their respective neonates delivered at the labour wards of University of Uyo Teaching Hospital (representing the tertiary healthcare institution) and St. Lukes Hospital, Anua (representing secondary healthcare institution), whose consent has been obtained to participate in the research. A capital letter "C" was written on the folders of mothers that gave their consent to participate in the research from antenatal ward.

Sample Size Determination

Calculation of minimum sample size was done using the formula proposed by Gorstein *et al.*[12] and based on the prevalence of congenital malaria of 9.6% reported in a multicentre study conducted in Calabar by Oduwale *et al*[13]. Provision was made for 10% attrition rate from incomplete documentation and 5% margin of sample error. A total of 220 mother/baby pairs who consented to participate in the study were recruited, between January to July, 2013, of which 218 mother/baby pairs information were actually available with complete information and data subsequently analysed.

Ethical Clearance

Ethical clearance was obtained from the Ethical Review Committee of the two hospitals, while informed consent was sought and obtained from all the subjects or their guardians before recruiting them in the study.

Data Collection

A pre-treated questionnaire was issued by the medical staff of the labour wards to obtain information 1 hour to 2 hours after delivery on demographic data, gravity and gestational age of mothers. The weight and gender of neonates were recorded. The questionnaires were issued to only mothers who had given consent to participate in the research. Data such as the blood group, age, incidence of malaria, the intermittent preventive measures taken during pregnancy and gravidity were obtained from mothers through the issued questionnaires. Aseptic procedures were observed in sample collection immediately after delivery.

Sample Collection

Medical staff of the labour ward collected cord blood as soon as new-born babies were separated from their mothers; the umbilical cord attached to the baby was clamped and released slightly to drop some blood into ethylene diamintetracetic acid (EDTA) bottles and labelled. They also obtained placental samples with sterile needles and syringes from the maternal placental surface into EDTA sample bottles and labelled. Samples from the babies were not collected for histology or placental impression smears. Capillary blood samples were collected from mothers (finger prick) and babies (heel prick) within 24 hours of delivery. Full blood counts (FBCs) were determined with an automation cell Dyn 3200. Women with haemoglobin (Hb)<11 and >7g/dl were classified respectively as anaemic and severely anaemic. A thick and thin blood smear of the same blood sample was made on a slide to determine the presence and species of *Plasmodium* .

Laboratory Analysis

This was carried out at University of Uyo Teaching Hospital Laboratory, Uyo. A thick and thin blood smear of the same blood sample was made on a clean grease free microscope slide, using a small drop of blood added to the centre of the slide and a larger drop about 15mm to the right. The drop of blood was immediately spread using a smooth edge slide spreader to make a thin film. Without delay, the large drop of blood was also spread to make a thick smear. The blood was allowed to air-dry with the slide placed in a horizontal position on a rack. A Giemsa stain diluted by adding 5ml of it to 45ml of buffered

water (or saline), pH 7.1-7.2 and mixed gently to obtain a 10% solution for 10minutes was used to stain the film on the slide for 10minutes. The slide was washed using clean water and the back of each slide was wiped clean and placed on a draining rack to air dry. The WHO recommended 7x eye pieces which gives a brighter and clearer image as against the 10x oil immersion objective lens was used for microscopic examination of the stained slides.

Data Analysis

Data generated from this study were analysed using independent sample t-test for equality of means and Chi-square for measurement of association among categorical variable at 95% significant level, at p-values less than 0.05($p<0.05$) were considered statistically significant.

RESULTS

A total of 220 mother/baby pairs were recruited into the study. Out of these, 218 mother/baby pairs consented to participate, and information obtained from them was subsequently analysed. Mother/baby pairs from University of Uyo Teaching Hospital (UUTH) was 140(65%), while 76(35%) was from St. Lukes Hospital, Anua.

Table 1 shows the age bracket of the participating mothers. Majority of the women who participated were between the ages of 27-33years, 117(53.7%); while those aged 20-26, were 79(36.2%). Those aged 34 and above were 14(6.4%) and only 8(3.7%) were below 20 years.

Table 1: Age of Mothers

Age Bracket (yr)	Total (%)
27-33	117 (53.7%)
20-26	79(36.2%)
Above> 34	14(6.4%)
Below< 20	8(3.7)

Table 2 shows the prevalence of malaria during pregnancy. Out of a total of 152 (69.7%) pregnant mothers: 99 from UUTH and 53 from St. Lukes hospital, Anua; pregnant mothers in third trimester had the highest episode of clinical malaria with 50(23.0%) prevalence rate, followed by those in second trimester with 45(20.6%) and those in first trimester with 43(19.7%), while the least prevalence of malaria was seen among mothers in more than one trimesters with 14(6.4%) only.

Table 2: Prevalence of Malaria During Pregnancy

Trimesters	UUTH	St. Luke	Total (%)
First	28(65.1)	15(34.9)	43(19.7)
Second	30(66.7)	15(32.3)	45(20.6)
Third	34(68.0)	16(32.0)	50(23.0)
More than one	9(64.3)	4(35.7)	14(6.4)

N = 152

Table 3 shows the different preventive measures taken by participanting mothers. Of the 218 participating mothers, 141(64.5%) participant took intermittent preventive treatment with sulphadoxine pyrimethamine (IPT-SP), 21(9.4%) used insecticide treated bed nets (ITN) and 15(6.9%) combined both IPT SP and ITN. However, 53(24.5%) did not take any prophylactic measures against malaria during pregnancy.

Table 3: Preventive measures among Participating Mothers

Preventive measures	UUTH	St. Luke	Total (%)
IPT-SP	92(65.3)	49(34.7)	141(64.5)
ITN	14(66.7)	7(33.3)	21(9.4)
IPT-SP/ITN	10(66.7)	5(33.3)	15(6.9)
No preventive measures	34(64.2)	19(35.8)	53(24.5)

Table 4 shows the different term of participating mothers. Majority of the babies (202) were born a term while 16 babies were pre-term. Four (4) of the babies (14.3%) were found to have malaria parasite among the pre-term, while 24(85.7%) were found in the term deliveries.

Table 4: Gestational Age of Participating Mothers at Delivery

Delivery term	UUTH (%)	St. Luke (%)	Total (%)	Malaria(+ve) %
	131(64.9)	71(35.1)	202(35.1)	24(85.7)
	11(68.8)	5(31.2)	16(7.3)	4(143)

Table 5 shows the prevalence rate of malaria

Table 5: Prevalence rate of malaria parasitaemia in maternal, neonatal, placental and cord blood.

Variables	(+ve) mp %	UUTH (%)	St. Luke (%)	Mean parasitaemia density
Maternal	8(3.7)	5(62.5)	3(37.5)	227.43± 0.55
Neonatal	28(12.8)	18(64.5)	10(35.7)	180.46± 0.33
Placental	25(11.5)	16(64.0)	9(36.0)	533.81± 1.50
Cord blood	27(12.4)	18(66.7)	9(32.3)	241.25± 0.89

Key: mp +ve = malaria positive

parasitaemia in maternal, neonatal, placental and cord blood to be 8(3.7%), 28(12.8%), 25(11.5%) and 27(12.4%) respectively, with neonatal blood samples having the highest rate. The mean parasitaemia density of 533.81±1.50 was found to highest in placental compared to other categorical variables. Others were; cord blood 241.25±0.89, maternal 227.43±0.55 and neonatal 180.46±0.33.

DISCUSSION

Table 6: Relationship between genotype and malaria parasitaemia

Genotype	Maternal blood mp +ve (%)	Neonatal blood mp +ve(%)	Placental blood mp +ve (%)	Cord blood mp +ve (%)
AA	6(3.6)	21(12.4)	18(10.7)	21 (12.4)
AS	2(4.3)	7(14.9)	7(14.9)	6(12.8)
p-Value	0.82	0.66	0.42	0.95

Key: mp +ve = malaria positive

Table 6 shows the relationship between genotype and malaria parasitaemia. Statistical analysis using Chi-square shows a higher rate of parasitaemia among AA genotype than AS in the various blood samples (maternal, neonatal, placental and cord blood). Invariably, there is no strong evidence against independent relationship with p-values <0.05. This may suggest that the higher prevalence in AA genotype is likely an occurrence due to chance.

Table 7: Relationship between congenital, maternal, placental and cord blood parasitaemia

Co-variant	mp +ve	X^2	Neonatal blood p-Value	Parasitaemia remark
Maternal	0(0%)	0.32	0.57	Not significant
Cord blood	2(7.1%)	0.35	0.37	Not significant
Placenta	9(32.1%)	13.53	0.00	Statistically Significant

Key: mp(+ve) = malaria positive

Table 7 shows the relationship between congenital, maternal, placental and cord blood parasitaemia. Out of the 8 women with malaria parasitaemia, none gave birth to malaria positive babies. While 27 parasitaemic cord blood samples had two babies pair who tested positive to congenital malaria. The Chi-square test analysis shows that a statistical significant relationship exist between placenta blood parasitaemia and neonatal blood parasitaemia (congenital malaria) with p-value <0.05.

Congenital malaria can be defined as the malaria infection resulting from the transplacental transmission of malaria parasite in maternal blood (particularly, *Plasmodium falciparium*), to the new born during pregnancy or prenatally during labour[1]. The prevalence rate of congenital malaria observed in this study was 12.8%. In Tanzania prevalence of congenital malaria was 19.1%[14]. Evidence from most of the cross sectional studies conducted in parts of sub-saharan African on congenital malaria within the last two decades (1990-2010) showed that congenital malaria is not uncommon as previously thought[15]. In Calabar, Nigeria, a prevalence of 2.0% was reported using Polymerase Chain Reaction (PCR)[16]. However, reports from other parts of Nigeria indicate prevalence rates ranging from 5.1% to as high as 54.2%[16]. This study revealed that the prevalence rate of parasitaemia in umbilical blood was 12.1%. However, the presence of malaria parasites in umbilical cord blood as indication of infection acquired ante-natally or as a result of contamination with infected maternal blood at delivery as stated in recent studies is still unclear[17]. In 2006, Malhotra *et al* [18] demonstrated from their study in Kenya that malaria parasites identified in cord blood were acquired ante-natally with a prevalence rate of 19.1%.

This study revealed that the overall prevalence of placental malaria among patients in the study population was 11.5% comparable to the prevalence of 10.3% and 10.5% obtained by a study carried out in Calabar[18] and Ibadan[16] respectively. This finding has confirmed that placental malaria is a constant feature in pregnancy in areas where malaria is endemic. Also comparing this association between congenital malaria and maternal, cord blood and placental parasitaemia as obtained from the study, placental blood parasitaemia showed the highest prevalence 9(32.1%) of congenital malaria. The chi-square test analysis also showed that there is a statistical significant relationship between placenta blood parasitaemia and neonatal blood parasitaemia (congenital malaria) with p-value less than 0.05. Therefore, null hypothesis that there is a significant relationship between placental parasitaemia and congenital malaria is accepted. This result when compared to the study in Calabar by Inyang-Etoh *et al.*[19] revealed a prevalence rate of 10.6% among patients who took IPT (Intermittent preventive treatment), while that among none IPT (Intermittent preventive treatment) patients was 11.3% (p=0.76). However, their study also revealed that there was no statistical significant difference in prevalence between the two groups. This study also revealed that although IPT reduces the degree of placental parasitaemia, it does not completely eradicate placental malaria. It also reminiscent the fact that current malaria control strategies are targeted at disease control rather than disease eradication. The result from this study supports the fact that the current malaria control strategies are targeted at control rather than eradication, because the information obtained from the participating mothers proved that 40% of women who were pre-treated with malaria prophylactic still tested positive to placental parasitaemia.

CONCLUSION

Congenital malaria is not uncommon in Uyo, Nigeria because a prevalence rate of 12.8% was obtained during the study. It is important that blood smears from neonates are taken and examined for the presence of malaria parasite soon after birth to ensure early treatment. Malaria prevention such as intermittent prevention treatment, prompt management of all malaria cases and use of insecticides treated bed nets should be reinforced for all pregnant women. *Plasmodium falciparum* was the only species encountered during this study. To curtail the incidence of acquiring congenital malaria, there is need for prompt and accurate diagnosis, and adequate antenatal care geared towards effective management and control of malaria.

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