Emerging Problems in Infectious Diseases

Plasmodium falciparum infection among neonates in the North Central Region of Nigeria

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Abstract

Introduction: This cross-sectional study investigated the rate of congenital and neonatal malaria infections in patients attending our hospital.
Methodology: Thick and thin blood films of 288 neonates admitted in the Special Care Baby Unit of Jos University Teaching Hospital, Nigeria, were examined microscopically for malaria parasites. Babies' and mothers' demographic and clinical data were analyzed.
Results: Of 288 blood samples examined, 160 (55.6%) were from males, 115 (39.9%) were from babies 0 to 7 days old, and 173 (60.1%) were from babies 8 to 28 days old. In total, 91 (31.6%) babies had malaria parasitaemia, of whom 49 (53.8%) were males. Malaria was significantly higher in babies 8 to 28 days old ($p<0.001$) and was independent of gender ($p=0.692$). Prevalence rates for congenital and neonatal malaria were 6.9% and 24.7% respectively.
Clinical presentations on admission included fever, cough, pallor, jaundice, and inability to suck. A total of 145 (50.3%) babies had symptoms of malaria, of whom 56 (61.5%) had malaria parasitaemia. Symptoms of malaria were present in 35 (12.2%) babies of 59 (20.5%) mothers who had symptoms of malaria during pregnancy. Ten (11.0%) of these neonates had malaria parasitaemia, of whom 4 (0.4%) were 0 to -7days old. *Plasmodium falciparum* was the only specie identified. No mortality was recorded against malaria-infected babies.
Conclusion: High prevalence of malaria in these neonates calls for high index of suspicion. Inclusion of malaria parasite test in the routine battery of tests for babies presenting with clinical signs and symptoms of neonatal infections is recommended.

Key words: *Plasmodium falciparum*; congenital and neonatal malaria; seasonal variation; high prevalence; Jos; Nigeria

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Introduction

Malaria continues to be one of the most important public health problems worldwide, and is responsible for 8% of the mortality among children below the age of five [1]. *Plasmodium* species are usually transmitted by anopheles specie mosquitoes, but can also be transmitted in other ways, including from mother to child causing congenital malaria [2]. Neonatal malaria was once thought to be rare, especially in malaria holo-endemic areas [3]. This was thought to be due to the protective effect of maternal antibodies that are passed to the newborn [4-6]and to the protective role of fetal hemoglobin in slowing the rate of the parasite development [7].

However, recent reports indicate that this notion may no longer be valid [8-9]. Sowunmi and colleagues [10] reported an incidence of 22.2% (n = 16) among 72 deliveries screened in Ibadan, South-West Nigeria. In a multi-center study, Falade *et al*. [11] noted that malaria incidence in Nigeria varied with geographical locations and additionally reported an overall incidence of 5.1% in the country. The authors had an incidence of 1.1% (n = 1,875) in Ibadan and 2.2% in Ilorin, North-Central Nigeria. High prevalence of congenital and neonatal parasitaemia (> 20%) has also been previously reported in Uganda and Zambia [12-13]. In Malawi, Kamwendo and colleagues [14] reported a prevalence of 6% malaria parasitaemia in cord blood by microscopy and 20% by polymerase chain reaction (PCR).

This study was conducted in the research laboratory of the Special Care Baby Unit of Jos University Teaching Hospital, Jos, Nigeria, from 1 July 2006 to 30 June 2008 to determine the rate of malaria infections among neonates admitted in the hospital. Malaria transmission in Nigeria occurs year-round with a major peak during the rainy season. *P. faciparum* infection, which is the most common in sub-Sahara Africa, is the most fatal. Jos is the capital city of Plateau state located in the Guinea Savanna belt of North-Central Nigeria It is characterized by its cool climate with two seasonal variations, the rainy season which occurs from April to October, and the dry season which occurs from November to April. The

temperature ranges from 13.55°C to 33.0°C and the rainfall is 0.0 to 741.0mm/month. The knowledge gained from this study will help in proper management of babies presenting with clinical signs and symptoms of neonatal infections.

Methodology

This cross-sectional study was conducted at Jos University Teaching Hospital, Jos, Nigeria. The hospital receives referrals from different parts of Plateau state and its neighbouring states.

Study population and methodology

The study population consisted of 288 neonates managed in the Special Care Baby Unit of Jos University Teaching Hospital. Blood samples sent to the unit's research laboratory for various routine investigations from 1 July 2006 to 30 June 2008, were also analyzed for malaria parasite.

Thick and thin blood films of respective specimens were made from an EDTA anti-coagulated blood less than one hour after the blood was drawn. Blood films were air-dried without convection for one hour and stained with 10% freshly prepared Giemsa stain. Thin blood films were fixed with 100% methanol prior to staining. Quality-controlled Giemsa stain, dust-free microscopy glass slides and phosphate buffer pH 7.2 were used.

Giemsa-stained thick and thin blood films were examined for malaria parasite using an oil immersion objective microscope (x100) by an experienced medical microbiologist in the laboratory. Malaria diagnosis was based on identification of asexual stages of *Plasmodium* on the thick blood smears. Film was reported as "malaria parasite not seen" (*i.e.*, negative) after examining about 100 fields. Thin films were used to identify species and stages of *Plasmodium* or other blood-borne pathogens [15]. Parasite density was measured by the number of parasites per microlitre of blood (thick film) [15]. The number of asexual parasitic forms (trophozoites and schizonts) present in as many microscopic fields as possible necessary to count 200 leucocytes was recorded. The standard value of 8,000 WBC/µl was assumed as a multiplier in the parasitaemia expression below:

$$\text{parasite/µl of blood (parasite density)} = \frac{\text{N x total WBC counts/µl (8000)}}{\text{leucocyte count (200)}}$$

where N = the number of asexual parasitic forms present in as many microscopic fields as possible to count 200 leucocytes.

Runsewe-Abiodun and colleagues' definition of congenital malaria as symptoms attributable to malaria only with evidence of intra-erythrocytic asexual forms of *Plasmodium* species in the first seven days of life, and neonatal malaria as intra-erythrocytic asexual forms of *Plasmodium* species in the first 28 days of life in a sick neonate [3], was used to define congenital and neonatal malaria respectively. Any neonate requiring hospitalization was considered "sick" [16]. Babies aged 0 to 7days and 8 to 28 days with malaria infection were grouped as congenital malaria and neonatal malaria groups respectively.

Extraction of data from the patients' hospital records was performed retrospectively. Members of the research team in conjunction with staff of the Records department of the hospital collected the hospital records. Data extracted from the hospital records included clinical presentations, outcome, sex, gender, temperature as on the day of analysis, birth weight and gestational age of the babies, as well as some of the mothers' ante-natal data which included age, signs and symptoms of malaria during pregnancy, and antenatal clinic visit status. Fever was defined as temperature greater than 37.5°C. Logistic regression model was used to determine the effect of parasite density on temperature while Pearson chi-square test was used to test for level of significance. All statistical analyses were performed using Epi Info version 3.3.2 (http://www.cdc.gov/epiinfo/). Test of significance was put at values less than 5%.

Ethical considerations

Consent was sought and obtained from the Head of Department of Paediatrics before screening the routine blood specimens for malaria, and from the Head of the Records Department of the hospital before extracting data from the patients' hospital records.

Results

Table 1 shows the presence of malaria parasitaemia by the babies' ages and gender. Out of a total of 288 neonates involved in the study, 160 (55.6%) of the babies were males. One hundred and fifteen (39.9%) of the babies were between 0 and 7 days old, and 173 (60.1%) were between 8 and 28 days old. Ninety-one (31.6%) babies had malaria parasitaemia. The mean age of the babies aged 0 to7 days was 4.27 ± 2.21 days, while that of the babies aged 8 to 28 days was 14.13 ± 5.23days. The mean age of the mothers was 27.40 ± 6.36years. The mean temperature for the babies with fever was 37.70 ± 0.80^{o}C. The prevalence rates for congenital and

neonatal malaria were 6.9% and 24.7%, respectively. Malaria infection was significantly higher in babies within 8 to 28days old ($p < 0.001$). There was no significant difference in the rate of malaria infection by gender ($p = 0.692$). *P. falciparum* trophozoites (ring forms) were the only specie and stage identified.

Two hundred (69.4%) of the mothers completed all their antenatal visits and care. Some of the clinical presentations by the babies on admission included fever, jaundice, anaemia, cough, and refusal to suck. All (100%) of the babies with symptoms of malaria had fever (temperature > 37.5^{o}C). Fifty-six (61.5%) babies out of the 145 (50.3%) who were symptomatic for malaria had malaria parasitaemia (Table 2). Symptoms of malaria were present in 35 (12.2%) babies of 59 (20.5%) mothers who reported symptoms of malaria during pregnancy; 10 (11.0%) of these neonates had malaria parasitaemia, 4 (0.4%) being within 0to 7 days old. There was no mortality recorded against any of the malaria-infected neonates (Table 2).

Table 3 shows the parasite density levels by clinical presentations of the babies. Parasite density ranged from 01 to 400 parasites/µl. Most of the babies had low parasitaemia. Babies presenting with fever, inability to suck, and jaundice had the highest levels of parasitaemia.

Gestational age and birth weight categories of the babies as well as the association between malaria parasitaemia and birth weight had been reported by the authors in their previous studies [17-18]. In the present study, the mean gestational age of babies with or without congenital malaria was 34.96 ± 4.18 weeks

Table 1. Malaria parasitaemia by babies' age and gender

	parasitaemia				
	MP not seen	MP seen (%)	Total (%)	Age (days)	p-value
0-7days	95 (33.0%)	20 (6.9)	115 (39.9)	4.27 ± 2.21	0.000
8-28days	102 (35.4%)	71 (24.7)	173 (60.1)	14.13 ± 5.23	
Male	111 (38.5%)	49 (17.0)	160 (55.6)	-	0.692
Female	86 (29.9%)	42 (14.6)	128 (44.4)	-	

MP = malaria parasite

Table 2. Symptomatic and asymptomatic malaria by malaria parasitaemia

	1	2	3	4	5	Total
Mothers (%)	59 (20.5)	229 (79.5)	-	-	-	288 (100.0)
Babies (%)	-	-	35 (12.2)	110 (38.2)	143 (49.7)	288 (100.0)
MP not seen (%)	-	-	25 (12.7)	64 (32.5)	108 (54.8)	197 (100.0)
MP (*P. f*) seen (%)	-	-	10 (11.0)	46 (50.5)	35 (38.5)	91 (100.0)

1 = Symptoms of malaria during pregnancy
2 = No symptom of malaria during pregnancy
3 = Symptoms of malaria in babies whose mothers had symptoms of malaria during pregnancy
4 = Symptoms of malaria in babies whose mothers hadn't symptoms of malaria during pregnancy
5 = Babies without the symptoms of malaria
P. f = Plasmodium falciparum

Table 3. Parasite density levels by clinical presentations

Clinical presentations	MP not seen	MP seen	parasite density (parasites/µl)			
			01-100	101-200	201-300	301-400
1	32 (11.1%)	13 (4.5%)	11	2	-	-
2	78 (27.1%)	32 (11.1%)	24	6	2	-
3	28 (9.7%)	10 (3.5%)	7	3	-	-
4	14 (4.9%)	5 (1.7%)	3	2	-	-
5	31 (10.8%)	8 (2.5%)	8	-	-	-
6	6 (2.1%)	19 (6.6%)	4	13	1	1
Others	8 (2.8%)	4 (1.4%)	3	1	-	-
Total	197 (68.4%)	91 (31.6%)	60	27	3	1

1 = Jaundice + anaemia
2 = Fever + risk of sepsis + anaemia
3 = Low birth weight
4 = Fever + cough + excessive crying
5 = Cough + inability to suck
6 = Fever + inability to suck + jaundice

and 36.20 ± 4.71 weeks, respectively, while that for neonatal malaria was 36.17 ± 3.87 weeks and 36.64 ± 3.83 weeks, respectively. There was no significant difference between the gestational age of babies with or without congenital ($p = 0.257$) or neonatal ($p = 0.459$) malaria.

The logistic regression model of parasite density with temperature is shown in Table 4. Parasite density is a significant predictor of temperature ($p < 0.001$). There was a significant increase in temperature with every 0.0081 parasites/µl rise in parasite density (Figure 1).

Figure 2 shows the prevalence of malaria infection by seasonal (monthly) variation. Rate of malaria infection was higher during the rainy season (April to October).

Discussion

This study identified a prevalence rate of 6.9% for diagnosed congenital malaria and 24.7% for neonatal malaria. This finding is in agreement with those of an African survey on congenital malaria that had reported a mean prevalence rate of 7.0% and a range of 0% to 23% [19]. In Lagos, South-West Nigeria, Mukhtar *et al*. [9] reported an incidence rate of 15.3% (16/104) for congenital malaria, while in Kenya, Mwaniki and colleagues [17] had a prevalence rate of < 0.5%.

The 6.9% (20/288) prevalence rate of congenital malaria from this study is higher than the rate of 2.8% (8/284) reported for Jos by Egwunyenga *et al*. in 1995 [20]. The reason for this discrepancy may not be conclusively explained presently. However, it may be that the neonates in this study were sick, unlike the neonates, who were not necessarily sick, in the study by the cited authors. It is also worth considering that although microscopic identification of *Plasmodium* species remains the gold standard for diagnosis of malaria, PCR may offer an attractive addition for confirmatory identification and diagnosis and should be explored in future studies. This technique may give a more accurate description of the burden of congenital and neonatal malaria [21].

The prevalence rate of neonatal malaria (24.7%) from this study agrees with the 24.8% prevalence reported by Runsewe-Abiodun *et al.* [3] in Ogun state, South-west Nigeria. High prevalence rates of congenital and neonatal malaria greater than 20% have been reported in Uganda and Zambia [12,13]; thus the previous notion that congenital and neonatal malaria are rare appears to be no longer valid in this environment. The occurrence of diagnosed congenital malaria in these babies could be attributed to the maternal antibodies passed to the newborns not being able to completely clear the parasites [9,22].

The predominance of *P. falciparum* in this study is in line with the results reported in previous studies. The low parasitaemia rate and zero mortality recorded against malaria-infected babies in this study, which have also been reported by some other authors [11,23], could confirm that maternal antibodies still convey some protection against parasite multiplication and progression of disease and so reduces the risk of severe malaria in this group of patients [24]. However, bearing in mind the potential fatality of the *P. falciparum* specie, there is a call for greater concern for early and accurate diagnosis as well as appropriate case management to avoid undue morbidity and potential mortality.

Furthermore, the association of fever as the main clinical presentation for malaria is in keeping with the World Health Organization (WHO) case definition for malaria in endemic areas [3]. Fever could also be

Table4. Logistic regression model of parasite density with temperature

Source	ss	df	MS	Coeff.	std. error	T	p>/t/	95%	C.I
Model	12.8735	1	12.87						
Residual	49.7403	88	.5652						
Total	62.6139	89	.7035						
Temperature ^{O}C									
Parasite- density				0.0081	0.0017				
Constant				36.5583	0.1817	201.15	0.000	36.1972	36.9195

Number of Obs = 91
F (1, 88) = 22, 78
Prob > F = 0.0000
R-squared = 0.2056
Adj. R-squared = 0.1966
Root MSE = 0.75182

Figure 1. Temperature versus parasite density in neonates

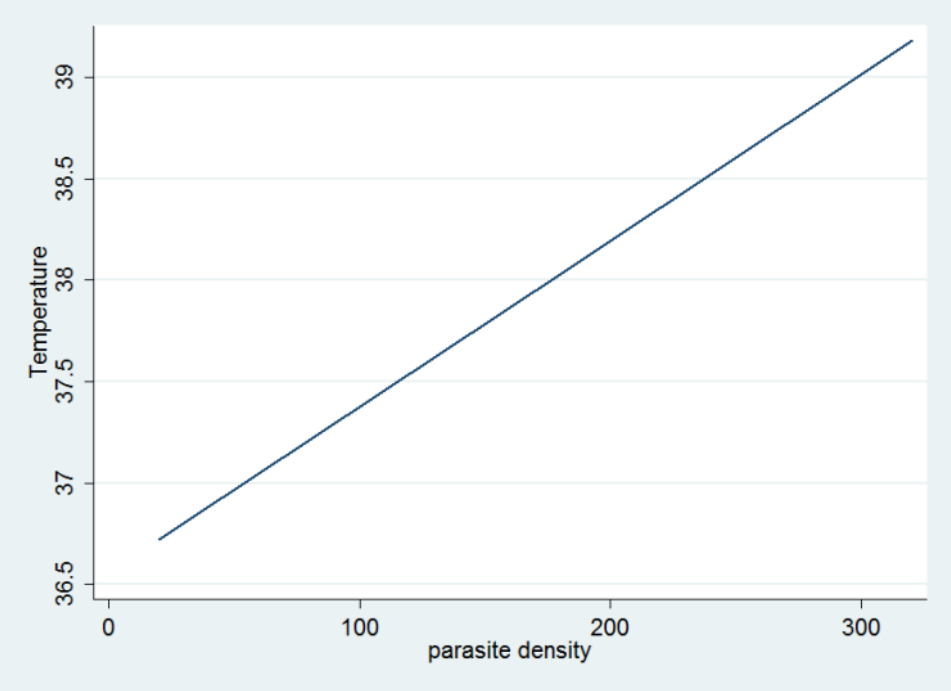
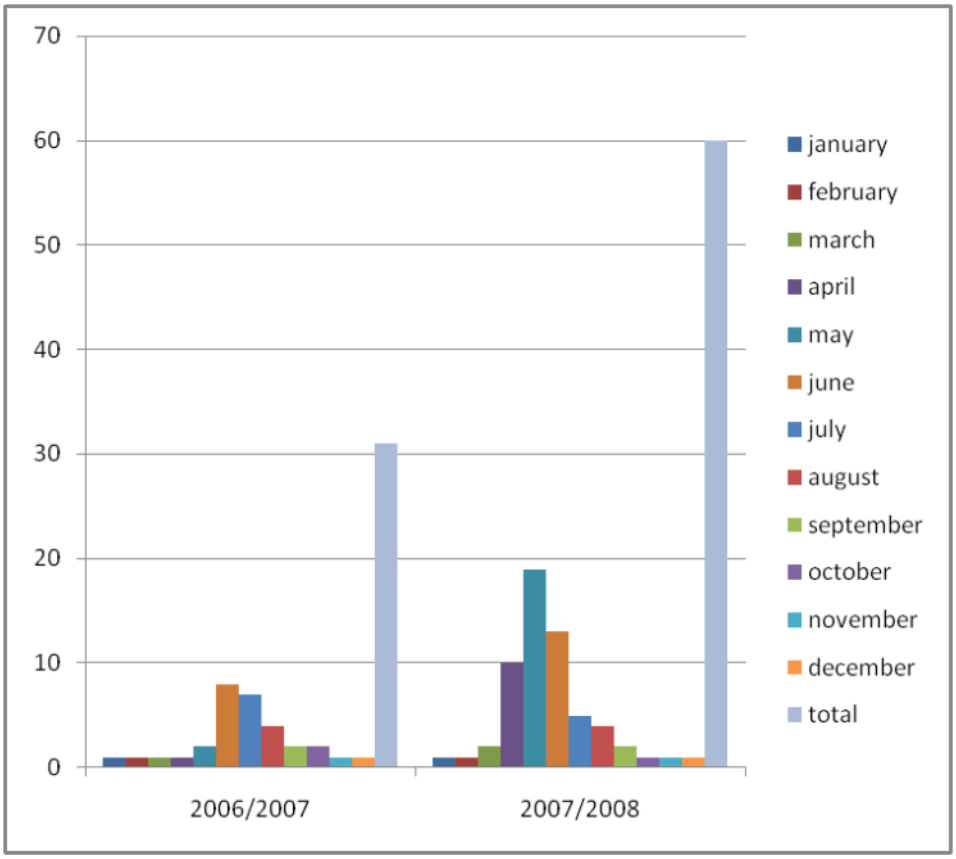


Figure 2. The prevalence of malaria infection by seasonal (monthly) variation



identified in some other neonatal infections. An earlier study reported that the signs and symptoms of malaria in newborns may be indistinguishable from those of other neonatal infections [3].Thus the importance of including the MP test as part of the battery of routine investigations for babies presenting with clinical signs and symptoms of neonatal infection cannot be overemphasized [25].

The presence of symptoms of malaria and diagnosed malaria in the neonates of mothers who had symptoms of malaria during their pregnancies implies that mothers who have symptoms of malaria during pregnancy should have a blood film test for malaria performed promptly and should be treated appropriately. Appropriate treatment of the mothers or absence of symptoms of malaria during pregnancy, however, does not obviate the need for heightened vigilance for symptoms of malaria in the offspring [24,26], as diagnosed congenital malaria infection was also seen in neonates whose mothers had no symptoms of malaria during pregnancy. Congenital malaria has been shown to occur in children of clinically healthy mothers who are delivered in malaria endemic-areas [27].

Parasite density is a significant positive predictor of temperature in children diagnosed of malaria. Although malaria parasitaemia occurred year-round, the prevalence of neonatal parasitaemia was highest during the rainy season when transmission was most intense. This observation implies the need for a high index of suspicion of malaria in babies presenting with clinical signs/symptoms of infections during this season.

Study limitations

One major limitation of our study was that the specimens studied were those sent to the unit's research laboratory within the stated time frame for various routine investigations, not necessarily the total number of admissions to the unit. Thus an additional investigation in which samples from the total number of admissions to the unit are studied may be necessary. Secondly, data extracted from the mothers' section of the babies' folders were based on the answers given by the mothers.

Conclusion

This study confirms that congenital and neonatal malaria are not as rare in North Central Nigeria as had previously been thought. There is, therefore, a call for a high index of suspicion for malaria and thus screening of neonates with presentations of neonatal infections for early, accurate diagnosis and appropriate intervention to prevent unnecessary morbidity and potential mortality.

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