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Research Article

Prevalence of Malaria Parasites among Pregnant Women and Children under Five years in Ekiti State, Southwest Nigeria

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Abstract

Background: Malaria is a deadly disease causing serious public health issues among pregnant women and children worldwide especially in tropical and subtropical Africa. This study was carried out to determine the prevalence of malaria parasites among pregnant women and children under five years in Ekiti State, Nigeria.

Methodology: A total of 380 blood samples were collected from the pregnant women and 100 children under five years respectively. Malaria parasites were examined microscopically on thick and thin blood smear stained with Giemsa stain while personal data were collected through questionnaire and confirmed from file records. Red cell phenotyping was carried out manually with standard tube technique for blood group. Haemoglobin electrophoresis was carried out using the cellulose acetate alkaline haemoglobin electrophoresis technique, which allowed for the separation of haemoglobin A, F, S, and C into distinct bands.

Results: The results showed that of 380 pregnant women sampled, 153 (40.2%) were positive for malaria parasites and 63 (63%) were positive of the 100 children sampled. The highest prevalence of malaria parasites 18 (51.4%) and 25 (71.4%) were observed in ages 36-39 and <1 years for pregnant women and children respectively. Multigravidae was 1.19 times (95% Cl: 0.77, 1.84) more vulnerable to malaria compare to primigravidae, but not significant. Women in the first trimester were more infected with malaria parasites 40 (75.4%) than those in second trimester 46 (23.3%) and third trimester 67 (51.9%). Among children under five years of age, females 38 (66.7%) had the highest prevalence compared to males 25 (58.1%). However, there was no significant difference. Statistical analysis showed a significant difference in genotype types ($P<0.05$).

Conclusion: This study revealed that malaria infection is still endemic in the study area, hence, there is urgent need to deploy management strategy to the study area.

Keywords: *Plasmodium falciparum*; Children; Pregnant women; Malaria parasite; Genotype; Blood group
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INTRODUCTION

Malaria is a major global health problem which poses risk to approximately 3.3 billion people in 97 countries and accounts for 214 million cases leading to about 600,000 deaths annually[1]. Malaria is a preventable and treatable infectious disease, which is transmitted through the bites of infected female *anopheles* mosquitoes.

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Malaria is caused by a protozoan parasite of the genus *Plasmodium*, four species namely; *Plasmodium vivax, P. ovale, P. malariae and P. falciparum* are responsible for human malaria[2]. P. falciparum accounts for about 80% morbidity and 90% mortality in humans [3], its common symptoms include headache, weakness, fever, pains, high body temperature (chills and rigors), bitter taste in the mouth and loss of appetite. In children, additional symptoms include more than sleeping, nausea and vomiting.

Malaria kills more than one million people every year, most of them in Sub Saharan Africa, where malaria

is a leading cause of death for children under five years and pregnant women [4]. In Sub Saharan Africa, malaria in pregnancy is predominantly asymptomatic and yet a major cause of severe maternal anaemia and low birth weight babies strongly associated with marked increase in infant mortality [5].

Malaria is endemic in Nigeria and its existence is well recognized and surveys reporting the prevalence in various communities in Nigeria [6-8]. Available records show that at least 50 percent of the population of Nigeria suffers from at least one episode of malaria each year [9]. High level of malaria endemicity, parasite resistance to affordable drugs and inadequate access to treatment facilities have contributed to making the disease the leading killer of children, accounting for an estimated 300,000 deaths each year. Also, many researchers have reported high prevalence rates of malaria in pregnancy in different parts of the country, ranging from 19.7% to 72.0% [10], with anaemia, pregnancy miscarriages and low birth weight of babies identified as the most debilitating effects of the disease which accounts for 11% of maternal deaths in the country [11].

In endemic areas, acquired immunity, though established is liable to break down the conditions of stress, in pregnant women. During pregnancy, there are usually high protein requirement and if dietary intake is insufficient, metabolic channels may be altered to withdraw protein from the immune system, hence the low the immunity in pregnant women [12]. Fetal and prenatal mortality, which sometimes lead to premature and false labour, occur in malarious mothers, although the incidence of preterm delivery is significantly increased only in non-immune mothers or those with low level of acquired immunity. Different studies have shown that malaria infection is more prevalent in primigravidae than in multigravidae [13].

Transmission of malaria is intense and stable in Nigeria because the infection remains constant throughout the year. The degree of endemicity of malaria measured is based on the spleen rate in children aged 2-9 years as published by WHO in order of severity. Hypoendemic malaria occurs when spleen rate in children is less than 10%, Mesoendemic occurs when the spleen rate is 11-50% in children, Hyperendemic occurs when spleen rate is 75% in children and greater than 25% in adults while Holoendemic occurs when spleen rate is greater than 75% in children but very low in adults. Malaria is holoendemic in Nigeria, with *Plasmodium falciparum* accounting for ninety five percent of all infections in the country [14]. Due to the deadly form of the malaria infection in the country, this present research was carried out at Ikole Specialist Hospital, Ekiti State, Southwest, Nigeria.

OBJECTIVES

- To determine the prevalence of malaria in pregnant women and children under five (5) years of age in the study area.
- To assess the stage of pregnancy in which the women are most susceptible to Malaria infection.
- To determine the relationship between haemoglobin genotype and blood group to Malaria parasite infection in the pregnant women and the children.

MATERIALS AND METHODS

Study Area and Population

Ikole Ekiti, the headquarters of Ikole Local Area of Ekiti state is located at longitude 4.50^0E and latitude 7.18^0N. Population of the area is about 168, 436 [15]. Ikole Ekiti has a tropical climate with lengthy and heavy rainy season which occurs between the months of April to October. The average temperature ranges between 25^0C and 28^0C in the year. The ecology of Ikole provides suitable breeding sites for biological multiplication, development and high survival rate of female *Anopheles* mosquito vectors for the transmission of malaria parasite to the populace.

This study was conducted at the Specialist Hospital, Ikole Ekiti, the only secondary and referral health facility in Ikole LGA of Ekiti state, South West, Nigeria. The hospital has a maternity ward for pregnant women. The facility carries out antenatal clinic (ANC) activities twice-weekly.

Data Collection

A structured interviewer-administered questionnaire was used to obtain information on socio-demographics and other factors related to malaria symptoms. These factors include age, occupation, educational status, regular use of ITNs, gestational age, and gravidity. Also, the file records were used for confirmation. The questionnaire was developed in English language using questions adopted from literature on related studies and also questions based on knowledge of the subjects by the investigators.

Three hundred and eighty (380) pregnant women and one hundred (100) children under five years of age who visited Ikole Specialist Hospital during the cross-sectional survey were involved.

Sample collection and Laboratory Analysis

About 2-3 ml of peripheral venous blood was aseptically collected from each participant into EDTA tubes by a trained laboratory technician. Thick and thin blood films were prepared on glass slides for parasite identification and speciation using Giemsa technique [16]. The slides were stained and viewed using x100 oil immersion objective lens. At least 100 high power fields were examined before a thick smear was reported as negative. Each slide was read independently by two trained microscopists and slides were reported as positive when both microscopists agreed on the reading.

Determination of ABO blood grouping

Red cell phenotyping was carried out manually with standard tube technique. For ABO blood grouping, a drop of anti-A, anti-B, and anti-AB (Biotec Laboratories Ltd, Ipswich, UK) was added into labelled clean test tubes containing a drop of the sampled blood. The contents were tapped gently to mix and centrifuged for 30 seconds at 1,000 rpm. The cell buttons were gently resuspended and observed for agglutination. Presence of agglutination

Table 1: General characteristics of the participating pregnant women against Malaria status (n=380)

	Malaria Status (%)				
Variables	**Positive**	**Negative**	**Total (%)**	χ^2	**P - Value**
Pregnancy Gravidity					
Primigravidae	48 (37.5)	80(62.5)	128 (33.68)	0.61	0.43
Multigravidae	105 (41.7)	147 (58.3)	252 (66.32)		
Trimester					
First	40 (75.5)	13 (24.5)	53 (13.95)	58.50	0.01
Second	46 (23.2)	152 (76.8)	198 (52.10)		
Third	67 (51.9)	62 (48.1)	129 (33.95)		
Age					
20 – 23	21(26.3)	59 (73.8)	80 (21.05)	14.59	0.01
24 – 27	29 (48.3)	31 (51.7)	60 (15.79)		
28 – 31	47 (46.1)	55 (53.9)	102 (26.84)		
32 – 35	31 (33.0)	63 (67.6)	94 (24.74)		
36 – 39	18 (51.4)	17 (48.6)	35 (9.21)		
40 – 43	2 (22.2)	7 (77.8)	9 (2.37)		
Blood Group					
A	23 (54.8)	19 (45.2)	42 (11.05)	40.78	0.01
B	37 (74.0)	13 (26.0)	50 (13.16)		
AB	15 (53.6)	13 (46.4)	28 (7.37)		
O	78 (30.0)	182 (70.0)	260 (68.42)		
Genotype					
AA	99 (51.0)	95 (49.0)	194 (51.05)	19.50	0.01
AS	53 (29.4)	127 (70.6)	180 (47.37)		
SS	1 (16.7)	5 (83.3)	6 (1.58)		

χ^2 – Chi Square Value, P value - Significance

constituted positive results, whereas absence of agglutination constituted negative results.

Determination of haemoglobin electrophoresis patterns

Haemoglobin electrophoresis was carried out using the cellulose acetate alkaline haemoglobin electrophoresis technique, which allowed for the separation of haemoglobin A, F, S, and C into distinct bands. Haemolysate of each sample was prepared and electrophoresed in a haemoglobin electrophoresis chamber containing Tris buffer solution for 20 minutes at 230 V. Haemolysate from blood samples of known haemoglobin types were run as a control. The result was read by comparing the distance of migration of the test sample with known controls.

Statistical Analysis

The data was analyzed using Chi-square and Binary Logistic Regression Model at 5% level of significance.

Ethical Clearance and Consent to Participate

This study was approved by the Ethical Research Committee of the Ekiti State Ministry of Health [Ref: ES/MOH/644/2014]. A written informed consent was obtained from all pregnant women prior to their enrolment in the study. Confidentiality of the participants and the information provided were assured and maintained throughout the study period.

RESULTS

A total of four hundred and eighty (480) individuals participated in the study; 380 (79.2%) were pregnant women while 100 (20.8%) were children.

Prevalence and distribution of malaria parasite among pregnant women

Among the pregnant women, Primigravidae were 128 (33.7%) while Multigravidae were 252 (66.3%). The pregnant women on their first trimester were 53 (13.9%), second trimester were 198 (52.1 %) while third trimester were 129 (33.9 %). Their genotypes include AA (51.1%), AS (47.4%) and SS (1.6%). Of the one hundred (100) children, 43% were males while 57% were females. Their genotypes were AA (32%), AS (60%), and SS (8%) (Table 1).

Out of the 380 blood samples collected from the pregnant women, 153 (40.2%) were positive for malaria infection. Of the 128 primigravidae sampled, 48(37.5%) were infected with malaria parasite while the highest prevalence of 105(41.7%) was obtained from 252 multigravidae sampled (Table 1). Although, Chi-square showed no significant difference ($\chi^2 = 0.61$, $p = 0.43$) in infection rate of the pregnancy gravidity (Table 1).

Table 2: Binary Logistic Regression Model on socio-demographic determinant on malaria in pregnant women

Variables	Variable Coefficient (B)	Unadjusted OR (95% Cl)	Significancy
Pregnancy Gravidity			
Primigravidae (Ref)	1.0		
Multigravidae	0.17	1.19 (0.77, 1.84)	Not significant
Trimester			
First (Ref)	1.0		
Second	-2.34	0.10* (0.05, 0.20)	
Third	-1.05	0.35* (0.17, 0.72)	
Age			
20 – 23 (ref)	1.0		
24 – 27	0.97	2.63* (1.29, 5.35)	
28 – 31	0.88	2.40* (1.23, 4.52)	
32 – 35	0.32	1.38 (0.72, 2.67)	Not significant
36 – 39	1.09	2.98* (1.30, 6.82)	
40 – 43	-0.22	0.80 (0.15, 4.17)	Not significant
Blood Group			
A	1.04	2.83* (1.46, 5.48)	
B	1.89	6.64* (3.35, 13.18)	
AB	0.99	2.69* (1.22, 5.92)	
O (ref)	1.0		
Genotype			
AA (ref)	1.0		
AS	-0.92	0.40* (0.26, 0.61)	
SS	-1.65	0.19 (0.02, 1.67)	Not significant

* p <0.05. Positive to Malaria status was coded 1 while negative was coded 0 in binary logistic regression.

Table 2 showed that multigravidae was 1.19 times (95% Cl: 0.77, 1.84) prone to malaria than the primigravidae, but not statistical significant. Regarding the gestation period, the highest malaria prevalence of 40 (75.5%) was significantly observed in the first trimesters (χ^2 = 58.50, p = 0.01) while the second trimester (OR= 0.10, Cl: 0.05, 0.20) and third trimester (OR= 0.35, Cl: 0.17, 0.72) were less vulnerable to malaria compared to those in their first trimester. Malaria infection seems to relatively decrease with increasing gestation period.

The malaria prevalence with respect to age among pregnant women reflected a galloping pattern. However, it was observed that the age group 36-39 years had the highest prevalence of 18 (51.4%)followed by age group 24-27years with 29 (48.3%) prevalence while the least was observed in age group 40-43 with prevalence rate of 2 (22.2%). Chi-square analysis revealed that there is a significant difference (χ^2 = 14.59, p = 0.01) in the age group.

Highest prevalence of malaria infection 99(51.0%) was observed in pregnant women with genotype AA, followed by genotype AS with 53(29.4) while those with genotype SS had the least prevalence 1(16.6). The vulnerability to malaria infection was less in SS (OR = 0.19 Cl: 0.02, 1.67) compared to AA genotype, but not significant. Chi-square test showed significant difference (χ^2 = 40.78, p = 0.01) in blood group of the pregnant women studied. B blood group had highest prevalence of (74.0%) while the least prevalence of malaria infection was observed in blood group O (30.0%). Blood group A was 2.83 times (Cl: 1.46, 5.48) prone to having malaria than O blood type.

Prevalence and distribution of malaria parasite among children under five years

Table 3 shows the prevalence of malaria parasite among pre-school children while Table 4 shows their probability of having malaria. Out of 100 children examined for malaria parasites, 63 (63%) were infected. No statistical significant in age difference (χ^2 = 4.34, p = 0.36) but prevalence of malaria parasite decreases with increase in age except for age 3 years with the least prevalence of 43.7% (Table 3). Age 5 was less likely (OR = 0.47 Cl: 0.13, 1.74) to have malaria compare to age 1 (Table 4), but not significant. Chi-square test showed no significant difference (χ^2 = 0.72, p = 0.38) in sex of the children, though the prevalence in females were 1.44 times (Cl: 0.64, 3.27) higher than males, but not significant. The degree of susceptibility of different genotype to malaria parasite as observed in the children sampled revealed significant difference (χ^2 = 25.066, p = 0.01). The vulnerability of AS (OR = 0.03 Cl: 0.01, 0.25) and (OR = 0.01 Cl: 0.01, 0.14) having malaria were lesser than AA genotype.

DISCUSSION

This study revealed that malaria parasite was prevalent in Ikole, Ekiti State. It was apparent that *P. falciparum* was the only species observed which also had been confirmed earlier as the predominant species in Sub Saharan Africa by [17]. Results showed that the prevalence of malaria varied considerably between ages, gravidity, trimester, genotypes and blood group of the pregnant women screened. This study revealed that a relatively high number of the pregnant women (40.2%) had

Table 3: General characteristics of the participating children against Malaria status (n=100)

Variables	Malaria Status (%)		Total (%)	χ^2	P-value
	Positive	Negative			
Age (years)					
0-1	25 (71.4)	10 (28.63)	35 (35)	4.34	0.36
2	13 (68.4)	6 (31.6)	19 (19)		
3	7 (43.8)	9 (56.3)	16 (16)		
4	11 (64.7)	6 (35.5)	17 (17)		
5	7 (53.8)	6 (46.2)	13 (13)		
Sex					
Male	25 (58.1)	18 (41.9)	43 (43)	0.77	0.38
Female	38 (66.72)	19 (33.3)	57 (57)		
Genotype					
AA	31 (96.9)	1 (3.1)	32 (32)	25.06	0.01
AS	30 (50.0)	30 (50.0)	60 (60)		
SS	2 (25.0)	6 (75.0)	8 (8)		

χ^2 – Chi Square Value, P value - Significance

detectable *P. falciparum* infection. This finding is slightly higher than those of Houmsou RS, et al[18] where he recorded 36.2% prevalence in studies carried out in Jos, Bauchi and Eku regions of Nigeria.

Multigravidae was more vulnerable to malaria infection than in primigravidae in this study, by having the highest prevalence, but not significant. This may be as a result of low level of specific immunity to malaria infection and the immunological changes in host during pregnancy. This result corroborates the works of [19] in Luanda Angola where the multigravidae had the highest prevalence of malaria infection. In relation to trimesters, the pregnant women in their first trimester were more prone to malaria infection. This result is in agreement with the works of [13] in Western Kenya and [20] in Nigeria, but disagrees with the report of [23] where it was found out that pregnant women in their second trimester were more prone (had the highest prevalence) to malaria infection.

The prevalence of malaria parasite with respect to age group in the pregnant women sampled revealed that all the age groups were infected but 36-39 age groups had the highest prevalence of malaria parasites while age group 40-43 were less vulnerable. This result is similar to that of [6] in Oshogbo, Nigeria but disagrees with the work of [21] in Anambra, Nigeria where they recorded the highest prevalence in age group less than 21 years.

In this study, participants of blood group O were more populated, followed closely by blood group B, blood group A and the least was group AB which is similar to the report of [22], who reported that the ratio of blood group O to other blood groups is higher in geographic region where malaria is endemic. Blood group B was more significantly infected than other blood groups, followed by group A, AB and the least infected was blood group O. This result also corroborates the study of [23] who reported that group B was the most vulnerable. This result might be due to the strong rosette formation with groups B RBCs which form rosette more than group O cells [24]. Most recently, it was confirmed that group A targets formed the strongest rosette.

In this study, 63% of the children population had malaria. The prevalence of malaria parasite observed in this research work was higher than 56.9% reported in a similar study in Jos, Nigeria by [25]. The variation in prevalence of malaria parasite among the children sampled could be attributed in part to the difference in

Table 4: Binary Logistic Regression Model on socio-demographic determinant on malaria in children

Variables	Variable coefficient (B)	Unadjusted OR (95% Cl)	Significancy
Age (years)			
0-1(ref)	1.0		
2	-0.14	0.87 (0.26 – 2.92)	
3	-1.17	0.31 (0.09 – 1.07)	Not significant
4	-0.31	0.73 (0.21 – 2.52)	
5	-0.76	0.47 (0.13 – 1.74)	
Sex			
Male (ref)	1.0		
Female	0.37	1.44 (0.64, 3.27)	Not significant
Genotype			
AA (ref)	1.0		
AS	-3.43	0.03* (0.01, 0.25)	
SS	-4.53	0.01* (0.01, 0.14)	

* $p < 0.05$. Positive to Malaria status was coded 1 while negative was coded 0 in binary logistic regression.

malaria transmission pattern, season of conducting the study and the use of malaria prevention tools. From this study, females were more vulnerable, however there was no significant difference ($p>0.05$) in gender. This result conforms to the result of [26] who reported higher prevalence of 57.9% in females than 42.1% in males. A predominance of malaria infection in males has been documented in some cases, but there is no scientific evidence to prove the higher prevalence being related to gender as susceptibility to malaria is not influenced by gender [27]. Also, studies have shown that females have better immunity to parasitic diseases which is attributable to genetic and hormonal factors [26]. In addition, age groups 1 year old had the highest malaria prevalence of 25 (71.4%) though there is no significant difference. This is in agreement with the report of [34] who reported the highest malaria prevalence (36.4%) in ½-2 age group.

Children with genotype AS and SS were significantly less vulnerable to malaria infection compared to genotype AA, although it is not significant. This result is similar to the results of [27] who reported 40.6% prevalence for genotype AA and SS (0%) and[6] who recorded 92.3% for genotype AA, AS (5.1%) and SS (2.6%) in Southern Nigeria.

CONCLUSION

Results of this study indicate that there is active transmission of malaria in the study area. The high prevalence observed, might be attributed to the period of study (May- September) which is the period of maximum rainfall in Nigeria.

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