

Original Research Article

***Plasmodium falciparum* parasitaemia during pregnancy and the use of malaria prevention methods by women attending antenatal consultation at the Regional Hospital Bamenda, Northwest Cameroon.**

ABSTRACT

Aims: The aims of this study were to determine the prevalence of *Plasmodium falciparum* parasitaemia in pregnant women and to identify the prevention methods used by pregnant women attending antenatal consultation (ANC) in Regional Hospital Bamenda (RHB).

Study design: This was a hospital based cross sectional study.

Place and duration of study: this study was carried out in the Regional Hospital Bamenda between April and May 2020.

Methodology: A structured questionnaire was used to collect information on sociodemographic characteristics, history of pregnancy and the use of malaria prevention methods. Capillary blood was collected through finger prick and used to prepare thick and thin blood smears that were Giemsa-stained for the detection of malaria parasite by microscopy.

Results: A total of 250 pregnant women took part in this study. The mean age \pm SD was 27.71 \pm 5.47 years. The mean pregnancy age \pm SD was 24.04 \pm 8.59 weeks. The prevalence of malaria parasitaemia was 18.0%. The most used malaria prevention method was LLINs (190/211, 91.0%). There was a statistically significant difference in MP prevalence between women who used IPTp (13.9%) and those who didn't (26.5%) ($\chi^2 = 5.87$, $P = .02$); between women who used bed nets and IPT (12.9%) in combination and those who didn't (24.0%) ($\chi^2 = 5.12$, $P = .03$) as well as between participants who used at least one method (16.4%) and their counterparts who didn't use any malaria prevention method (38.9%) ($\chi^2 = 5.73$, $P = .01$). Living in a house with plank or mud walls (13; 29.0%) was surprisingly more protective than living in a cement brick or mud brick house (30; 71.0%) (OR: 2.34; 95%CI: 1.10-4.97; $P = .03$).

Conclusion: Malaria remains a health preoccupation among pregnant women attending ANC consultation in the RHB. Use of bed nets and IPTp as well as the use of at least one prevention method could be protective.

Key words: *Plasmodium falciparum*, parasitaemia, prevention methods, pregnancy, antenatal consultation, Bamenda, Cameroon.

1. INTRODUCTION

Malaria during pregnancy is a significant public health problem with substantial risks for the pregnant woman, the foetus and the newborn child [1]. Several user friendly and efficient measures are recommended for the prevention of malaria to curb its morbidity and mortality.

Malaria-associated maternal illness, intrauterine growth retardation (IUGR), premature birth, low birth weight (LBW), neonatal death (NND), spontaneous abortion, stillbirth, maternal anaemia, are some of the complications associated with malaria in pregnancy (MIP) and are mostly the result of *Plasmodium falciparum* infection which is the predominant malaria parasite species in Africa according to WHO [1]. However, the severity of the disease varies with a number of factors including parity, pregnancy age, individual's level of acquired immunity as well as the implementation of prevention measures [2, 1, and 3].

In as much as all pregnant women are vulnerable to malaria parasite infection and disease, first time mothers (primigravidae) in high transmission settings are reported to be more at risk of infection with *P. falciparum* parasites than the multiparous [1]. Primigravidae and younger maternal age (adolescents) women are said to have poorer immunity to malaria compared to their multiparous and older counterparts and so are more exposed to infection [4, 5]. In most cases, malaria parasitaemia in pregnancy often presents as asymptomatic but progresses to symptomatic depending on the intensity of the infection, with higher parasitaemia progressing to the disease condition faster [6, 7]. This tendency has been associated with hormonal changes during pregnancy such as increase in cortisol which inhibits cell-mediated immunity against infected red blood cells. This causes sequestration of infected erythrocytes in the placenta as well as the expression of placenta parasite antigen on their cell membranes giving the immune system a second antigen to deal with [8].

Generally, the prevalence and severity of malaria infection tends to increase among pregnant women. In 2018, an estimated 11 million pregnant women were infected with malaria in areas of moderate and high disease transmission in sub-Saharan Africa. As a result, nearly 900 000 children were born with a low birthweight [9].

Plasmodium falciparum is the main malaria parasite in Cameroon with its endemicity varying in the different vegetation zones as a result of difference in the intensity of transmission. Northwest Cameroon is found in the humid savannah vegetation zone and is characterized by low intensity perennial transmission of the malaria parasite. Prevalence of malaria here has been reported to vary between 9.3 and 22.4% in children 1-15 years with an entomological infection rate (EIR) of 2.2-11.0 infective bites per person per year (ib/p/yr), justifying the endemicity of the parasite in the area [10]. Reports on malaria parasite prevalence among pregnant women have shown some level of variation. Sumo and collaborators reported 19.3% prevalence among pregnant women in Ndop, a locality in the same region as the study site [11]. Another study in Mamfe, Southwest Cameroon reported a prevalence of 39.6% [12]. No such study done in the Bamenda Regional hospital was found, reason why this study was necessary.

For effective prevention and control of malaria in pregnancy, the WHO recommends the use of long-lasting insecticide treated nets (LLINs) in all areas with moderate to high malaria transmission to prevent contact with the female *Anopheles* mosquito vector; intermittent preventive treatment in pregnancy (IPTp) with sulfadoxine-pyrimethamine (SP) from the second trimester to prevent adverse consequences of malaria on maternal and foetal outcomes; prompt diagnosis using reliable rapid diagnostic tests and microscopy; and effective treatment of malaria infections with artemisinin-based combination therapies in areas affected by resistance to monotherapy [1]. These measures have actually witnessed a scale-up in Cameroon [13].

The Cameroon government through the Ministry of Public Health and partner organizations has carried out the distribution of more than 20 million treated bed nets to households in the country since its adoption in 2010 [13]. Children under the age of five are treated free of charge for simple malaria while pregnant women are provided with LLINs during their first

antenatal visits in health facilities. In the same light intermittent preventive treatment using SP is given to pregnant women from second trimester of pregnancy during their ANC consultations since 2004 [14, 1].

Although resistance to SP has been reported, intermittent preventive treatment with SP during pregnancy (IPTp-SP) continues to provide significant benefit, particularly in the protection against both neonatal mortality and low birth weight even in areas where resistance to SP is reported [15, 16, 1 and 17]. This strategy and others cited earlier have generally contributed to curb the incidence of malaria parasite and disease among pregnant women [18, 19].

In line with prevention, individual and community based strategies taken towards fighting malaria are important contributors reported to have reduced malaria prevalence in several communities and countries especially when in combination with other WHO recommended measures [13]. Such measures including indoor residual spraying, use of insecticidal coils [20], the nature of houses [20, 21, and 22] and environmental sanitation [23], influence malaria transmission. Albeit the above cited measures, malaria remains endemic in Cameroon. In recent years, a declining effort to scale-up of IPTp in some African countries has been observed and countries are still to attain the 80% coverage target for at least 2 doses of IPTp recommended by WHO [9]. Hence malaria continues to be a public health concern especially among pregnant women.

In spite of its prevalence and endemicity, malaria remains a preventable disease. However, assessment of its prevalence as well as prevention measures among pregnant women seems uncommon, this leaves gaps in the information on prevalence and effectiveness of these measures in the country [24]. The assessment of such effectiveness and reporting could help in guiding policy and redefining strategies towards the fight against malaria in pregnancy. This study was aimed at determining the prevalence of *Plasmodium falciparum* parasitaemia in pregnant women and to identify the prevention methods used by pregnant women attending antenatal consultation (ANC) in the Regional Hospital Bamenda (RHB).

2. MATERIAL AND METHODS

2.1 Study site

This study was carried out at the Regional Hospital Bamenda, the headquarters of the North West Region of Cameroon during the months of April and May 2020. Recruitment of participants was at the ANC unit of the hospital. Bamenda is a multicultural and metropolitan town with inhabitants engaged in diverse socio-economic activities. The town has both public and private health facilities with a population of about 393,000 inhabitants in the urban area as of demographic trends [25]. It is located at Longitude 100.08 to 100.12 E and latitude 50.55 to 60.00 N, and about 1258-1770m above sea level. It has two seasons, a long rainy season from April to November and a short dry season which spans from December to March. With an average temperature of 23°C, March is the warmest month while July is the coldest with an average temperature of 20.1°C. The average annual rainfall is 2145 mm [26, 27]. The vegetation is mainly the savannah type. The town has a rough topography with hills and valleys.

2.2 Study design

This was a hospital based cross sectional study. Pregnant women attending antenatal consultation in the RHB were admitted into the study after obtaining their informed consent.

A structured questionnaire was used to collect information on sociodemographic characteristics, history of pregnancy and the use of malaria prevention methods. Blood samples were collected through finger prick and used to prepare thick and thin blood smears that were Giemsa-stained for the detection of malaria parasite by microscopy.

2.3 Sample size estimation and data collection

A sample size of 237 was determined using a standard formula [28] from the prevalence of MP in pregnant women in Ndop by Sumo and collaborators [11]. Data was collected from all pregnant women who gave informed consent during routine ANC visits. Explanation of the research procedures was done in the ANC lecture hall. Codes were attributed to each participant and written on the questionnaire and the participant's consultation booklet. Questionnaires were self-filled as the women waited to take rounds for their consultations. The questionnaires were in English and in French. Where necessary, the questions were verbally translated to Pidgin English. For those who could not read and write, the responses were filled by a member of the research team. Data on socio-demographics (age, level of education, profession) and history of the pregnancy (gravidity, gestational age and trimester) was obtained from the consultation booklet. Where data was unavailable, the women were asked directly. Trimesters were classified and defined as first (<14 weeks), second (14-26 weeks) and third (>27 weeks) [29]. The women were questioned on the use of malaria prevention measures including ownership and use of mosquito bed nets, use of IPTp, IRS, environmental sanitation, presence of ceiling and house type (walls made of cement bricks, mud bricks, plank or just mud). A blood specimen was then collected for white blood cell count and parasitological analysis.

2.4 Laboratory procedures

2.4.1 Blood specimen Collection, preparation and staining of thick and thin smears

A few drops of blood were collected through finger prick after filling the questionnaire. This was placed on a clean slide to prepare thick and thin blood smears. The slides were transferred to the Parasitology bench of the hospital laboratory for staining, determination of malaria parasite and quantification following standard measures. Twenty microliters (20 μ L) of the capillary blood was also collected to carry out a total white blood cell count using the Improved Neubauer ruled counting chamber. The thin blood smears were fixed with absolute methanol for a few seconds. Both blood smears were stained with 3% Giemsa for 30 minutes [30]. Thick smears were used to detect malaria parasites while the thin smears were used to identify *Plasmodium* species.

2.4.2 Speciation of malaria parasite

A binocular light microscope (Olympus, NY-USA) was used to examine the stained smears under the oil immersion objective (X100) and X10 eye piece. Each slide was read by two parasitologists, and where there was a disparity the reading of a third parasitologist was considered. The determination of *Plasmodium* species was confirmed using an identification chart by The Centre for Disease Control and Prevention [31].

2.5 Statistical analysis

Data collected was keyed into Excel version 2013 and analyzed using the SAS statistical software [32]. Results were presented as proportions which were compared using the chi square (χ^2) test while odds ratios (OR) from a Logistic regression model (LRM) was used to determine the physical risk factors of malaria parasitaemia. Both univariate and multivariate analysis were considered for the LRM. The level of significance was set at $P < .05$.

3. RESULTS AND DISCUSSION

3.1 RESULTS

3.1.1 Characteristics of the study population

A total of 250 pregnant women took part in the study. The age range was between 17-45 years with a mean (SD) of 27.71 (5.47) years. The majority 179 (71.6%) were ≤ 30 years. Eighty (32%) of them were primigravidae while 134 (53.6%) were in the third trimester. The overall malaria prevalence was 18% (45) as shown in Table. Only *Plasmodium falciparum* species was detected.

Table 1: Characteristics of the study population

Characteristic	Category	Frequency (N=250) (%)
Age(years)	Range	17.0 – 45.0
	Mean (\pm SD)	27.71 (5.47)
	≤ 30	179 (71.6)
	≥ 31	71 (28.4)
Educational level	Primary	50 (20.0)
	Secondary	92 (36.8)
	Tertiary	108 (43.2)
	Business	114 (45.6)
Occupation	Civil servant	32 (12.8)
	Farmer	15 (6.0)
	Other*	45 (18.0)
	Student	44 (17.6)
Gravida	First	80 (32.0)
	Second	69 (27.6)
	Third	65 (26.0)

Characteristic	Category	Frequency (N=250) (%)
Age of pregnancy (weeks)	>3	36 (14.4)
	Range	3.0 – 40.0
	Mean (\pm SD)	24.04 (8.59)
Trimester	First	39 (15.6)
	Second	77 (30.8)
	Third	134 (53.6)
Prevalence of malaria	Overall	45 (18)

**Other occupations including hair dressing, tailoring and mobile telephone call box attendant.*

3.1.2 Prevalence of malaria parasitaemia with respect to sociodemographic factors, gravida and trimester of pregnancy

Although malaria parasitaemia depicted a higher prevalence in women \leq 30 years (33, 73.3%), in business persons (27, 60.0%), a decreasing tendency with gravida and an increasing tendency with trimester, there was no statistically significant difference within any of these groups of participants as shown in Table 2.

Table 2: Prevalence of malaria parasitaemia according to sociodemographic factors, gravida and trimester of pregnancy.

Characteristic	Category	Prevalence of	
		MP (N=45) (%)	Chi-Square (P-value)
Age group	\leq 30	33 (73.3)	0.008
	\geq 31	12 (26.7)	(.78)
Occupation	Business	27 (60.0)	5.49 (.24)
	Civil servant	6 (13.3)	
	Farmer	2 (4.4)	
	Other*	5 (11.1)	
Educational level	Student	5 (11.1)	0.04
	Primary	9 (20.0)	

Characteristic	Category	Prevalence of MP (N=45) (%)	Chi-Square (P-value)	
Gravida	Secondary	16 (35.6)	(.98)	
	Tertiary	20 (44.4)		
	1	14 (31.1)	4.82	
	2	11 (24.4)		
	3	9 (20.0)		(.19)
	>3	11 (24.4)		
Trimester	First	9 (20.0)	4.08	
	Second	18 (40.0)		
	Third	18 (40.0)	(.13)	

**Other occupations including hair dressing, tailoring and mobile telephone call box attendant.*

3.1.3 Use of malaria prevention methods with respect to socio-demographic characteristics, **gravida and trimester of pregnancy**

Comparing the frequency of use of the different malaria prevention methods with respect to the socio-demographic characteristics and gravida did not reveal any statistical difference. However, a significant statistical difference ($\chi^2 = 66.96$, $P < .0001$) was observed in the uptake of IPT with respect to trimester, with the highest uptake being in the third trimester of pregnancy as shown in Table 3.

Table 3: Use of malaria prevention methods with respect to socio-demographic characteristics gravida and trimester of pregnancy

Characteristic	Category	Malaria prevention method (n, %)		
		Use of net (190, 91.0%)	IPT (165, 66%)	Sprays (62, 24.8%)
Age (years)	≤30	132	115	41

	≤31	58	50	21
χ^2 (P-value)		0.40 (.52)	0.68 (.41)	1.06 (.30)
Occupation	Business	84	66	27
	Civil servant	28	24	7
	Farmer	8	10	5
	Other	40	32	9
	Student	30	33	13
χ^2 (P-value)		6.23 (.18)	6.82 (.15)	1.58 (.82)
Educational level	Primary	38	35	11
	Secondary	66	56	23
	Tertiary	86	74	28
χ^2 (P-value)		0.09 (.95)	1.62 (.44)	0.46 (.84)
Gravida	1	61	53	15
	2	55	49	21
	3	48	42	16
	>3	26	21	10
χ^2 (P-value)		1.39 (.71)	1.74 (.63)	2.61 (.46)

	First (39)	24	8	10
Trimester	Second (77)	57	41	24
	Third (134)	109	116	28
χ^2 (P-value)		1.11 (.56)	66.96 (<.0001)	2.74 (.25)

3.1.4 Prevalence of malaria parasitaemia according to use of chemical prevention methods

The most used malaria prevention method was bed nets (91.0%) among those who possessed the nets, while the least used was indoor residual spraying (26.1%) Comparing malaria prevalence between those who used chemical prevention methods and those who didn't use revealed that there was a statistically significant difference ($\chi^2 = 5.87$, $P = .02$) between women who used IPTp (13.9%) and those who didn't (26.5%). Also, comparing malaria prevalence between women who used prevention methods in combination and those who didn't, a statistically significant difference ($\chi^2 = 5.12$, $P = .03$) in prevalence was observed only in women who used bed nets and IPT (12.9%) and those who did not (24.0%). Similarly, there was a statistically significant ($\chi^2 = 5.73$, $P = .01$) difference in prevalence between participants who didn't use any malaria prevention method (38.9%) and their counterparts who used at least one method (16.4%) as shown on Table 4.

Table 4: Prevalence of MP with respect to use of chemical prevention methods singly and in combination

Prevention method	Number tested	Category	Frequency	(%) Prevalence of MP (%)	of Chi-Square (P-value)
Use of net	211	No	21 (10.0)	4(19.0)	0.10 (.75)
		Yes	190 (91.0)	31(16.3)	
IPTp	248	No	83 (33.5)	22 (26.5)	5.87 (.02)
		Yes	165 (66.5)	23 (13.9)	

Prevention method	Number tested	Category	Frequency	(%) Prevalence	of Chi-Square
				MP (%)	(P-value)
Use of sprays	247	No	185 (74.9)	32 (17.3)	0.42 (.52)
		Yes	62 (25.1)	13 (21.0)	
Use of net + IPTp	249	No	117 (47.0)	28 (24.0)	5.12 (.03)
		Yes	132 (53.0)	17 (12.9)	
Use of net + sprays	248	No	208 (83.9)	36 (17.3)	0.61 (.44)
		Yes	40 (16.1)	9 (22.5)	
Use of IPTp + sprays	248	No	205 (82.7)	36 (17.6)	0.27 (.60)
		Yes	43 (17.3)	9 (21.0)	
Use of net + IPTp + sprays	249	No	219 (88.0)	39 (17.8)	0.15 (.70)
		Yes	30 (12.0)	6 (20.0)	
Use of at least one method	250	No	18 (7.2)	7 (38.9)	5.73 (.01)
		Yes	232 (92.8)	38 (16.4)	

3.1.5 Physical risk factors and malaria parasitaemia

In comparing malaria parasite prevalence among positive participants with respect to some physical risk factors, it was observed that there was a statistically significant difference (OR: 2.34; CI: 1.10-4.97; $P=.03$) between the prevalence of MP in participants living in cement brick and mud brick houses (30; 71.0%) and those living in houses with plank or unmolded-mud walls (13; 29.0%).

Table 5: Physical prevention methods with respect to malaria parasite positive participants

Prevention method	Category	Prevalence (N=45) (%)	OR (95% CI)	LRM P-value
Have net	No	10 (22.2)	2.56 (0.23-29.16)	.44
	Yes	35 (77.8)		
Frequency of net use	Always	25 (55.6)	0.71 (0.30-1.82)	.48
	Sometimes	7 (15.5)		
Type of house walls	Bricks ¹	32 (71.0)	2.34 (1.10-4.97)	.03
	Others ²	13 (29.0)		
Presence of ceiling	No	11 (24.4)	1.09 (0.43-2.73)	.86
	Yes	34 (75.6)		
Bushes near home	No	27 (60.0)	1.12 (0.52-2.40)	.77
	Yes	18 (40.0)		

1: walls made of cement and mud bricks; 2: walls made of plank or unmolded mud; LRM: Logistic regression model

3.2 Discussion

Prevention of malaria remains indispensable in the fight against malaria especially among vulnerable groups including pregnant women. The uptake of prevention methods has been shown to vary among pregnant women. However, given a scale-up of certain prevention measures such as mosquito bed nets in the population and IPT by governments including Cameroon, an increase in uptake of these measures is expected. In this regard, reporting on the use of these measures and the prevalence of malaria parasite provides requisite information for the assessment of the impact of these measures and probably a reorientation of used strategies.

The prevalence of malaria in this study (18.0%) is an indication that malaria remains endemic among pregnant women attending ANC in RHB. This prevalence is however lower than the national prevalence of 21% [19] and that obtained in Ndop [11] and Mamfe [12]. This lower prevalence in this study may be as a result of the uptake of the malaria prevention measures with time as well as variation in environmental conditions [21].

With respect to prevalence of malaria parasitaemia according to sociodemographic factors, gravida and trimester of pregnancy, it was observed that none of these factors showed a statistically significant impact despite some differences in prevalence. This may be as a result of the massive use of at least one method of prevention (92.8%) by the women leaving these characteristics with little possibility of influencing malaria prevalence.

Concerning prevalence of MP with respect to use of chemical prevention methods, the significantly lower malaria prevalence observed among participants who used IPTp compared to their counterparts who didn't use is an indication that IPTp continues to be protective against malaria parasite infection given that Sulphadoxine pyrimethamine has the ability to inhibit the reproduction of *Plasmodium* parasites [33]. The high uptake of bed nets and IPTp and the significantly lower malaria parasitaemia recorded among participants using IPTp and nets in combination than their counterparts who didn't use this combination is a possible indication of the effectiveness of these methods and the efficacy of the strategy put in place by health authorities [34]. In the same light, the significantly lower MP prevalence recorded among women who used at least one method of prevention compared to those who didn't use any method further corroborates this finding and affirms the protective role of prevention methods.

Among the chemical methods used, IRS was the least used. It was also observed that the prevalence of malaria was higher among women who used sprays whether singly or in combination compared to the other methods. This could be because the participants have to purchase them resulting to irregular use, but also, this could be related to the resistance to some insecticidal sprays reported in some areas [19].

Mosquito bed nets were the most used prevention method. The distribution of bed nets uses a strategy that meets the population at their door steps through quarter heads and community health workers. This increases the accessibility to the method compared to IPT which is at the level of the health facility and sometimes out of stock; and the sprays which must be purchased by the women. However, the highest uptake of IPT recorded in the third trimester could be because women delay in the start of ANC consultation or because as women get towards delivery they become more conscious and desire the protection of the pregnancy. But also, this could be because the distribution of IPTp in the health facility starts from the second trimester of pregnancy.

Among physical risk factors assessed in this study, it was observed that living in houses made of plank and unmolded mud walls was rather protective against malaria compared to living in houses made of cement and mud brick walls. This is contrary to reports from other studies [18, 21 and 22]. There is a possibility that living in houses with plank and mud walls could raise consciousness thereby pushing the individuals to seek other affordable measures of protection against malaria whereas, those in brick houses could be overconfident and neglect other prevention methods.

In this study, P-values were applied as a screening measure to get an indication of statistical strength but no correction for multiple testing was performed. Our study was also limited by missing data, particularly for the chemical and physical prevention methods, which was not accounted for in the analysis. Nevertheless, our results supported claims that prevention

methods are necessary during pregnancy. These findings might be useful for future studies on the assessment of the impact of malaria prevention methods on outcome of pregnancy.

4 CONCLUSION

Malaria remains a health preoccupation among pregnant women attending ANC consultation in the RHB. Use of bed nets and IPTp for prevention either singly or in combination as well as the use of at least one prevention method could be protective. The distribution of bed nets and IPTp intensified by health authorities may further reduce the prevalence of malaria among the women. Meanwhile all pregnant women should be encouraged to take up at least a malaria prevention method. Also, studies on the impact of malaria prevention methods on the outcome of pregnancy could add more information on the assessment of the current strategies.

CONSENT

Written informed consent was obtained from all women who took part in this study. All participants who were positive for malaria were referred to the consulting medical doctor for treatment. All participants consented that information collected in this study can be published.

ETHICS APPROVAL

An ethical clearance N° 2020/0118H/UBa/IRB was obtained from the Institutional Review Board of the University of Bamenda after reviewing the study protocol. Administrative authorization was obtained from the authorities of the Regional Hospital Bamenda.

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Abbreviations

ANC: Ante-natal clinic

CDC: Centres for Disease Control and Prevention

Ib/p/yr: Infective bites per person per year

IPTp: Intermittent preventive treatment in pregnancy

IRS: Indoor residual spraying

IUGR: Intra-uterine growth retardation

LBW: Low birth weight

LLINs: Long lasting insecticide treated nets

LRM: Logistic regression model

MIP: Malaria in pregnancy

MP: Malaria parasitaemia

NND: Neonatal death

OR: Odds ratio

RHB: Regional Hospital Bamenda

SD: Standard deviation

SP: Sulphadoxine Pyrimethamine

WHO: World Health Organization

X2: Chi square test

UNDER PEER REVIEW