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FACTORS ASSOCIATED WITH PLACENTAL MALARIA IN UPPER WEST REGIONAL HOSPITAL-WA, GHANA

PASCAL MWIN

(10325602)

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DECLARATION

I, Pascal Kingsley Mwin, declare that the result of this study is my own work conducted under supervision and I have duly cited and acknowledge other people's contribution that appear in the work. This research has not been wholly or partially presented for a degree in any other institution.

Date……. Signature…….

Principal Supervisor:

Dr. Frederick Wurapa

Signature……

Date: ……

Co-supervisor

Prof. Col. Edwin Afari (Rtd)

Signature……

Date: ……

DEDICATION

I dedicate this research to my parents for their immense support throughout my education.

ACKNOWLEDGEMENT

I am grateful to Almighty God for the blessing and giving me the strength to sail through the program despite the many challenges I faced.

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LIST OF ABBREVIATIONS

ANC	Antenatal Care
ERC	Ethical Review Committee
EDTA	Ethylenediaminetetraacetic Acid
GFELTP	Ghana Field Epidemiology & Laboratory Training Program
GHS	Ghana Health Service
HRP2	Histidine Rich Protein 2
IUGR	Intrauterine Growth Restriction
IPTp	Intermittent Preventive Treatment in Pregnancy
ITN	Insecticide Treated Bed Nets
IRS	Indoor Residual Spraying
LBW	Low Birth Weight
MiP	Malaria in Pregnancy
NMCP	National Malaria Control Program
OPD	Out Patient Department
PMI	Presidential Malaria Initiative
RDT	Rapid Diagnostic Test
SP	Sulphadoxine Pyrimethamine
WHO	World Health Organization
WIFA	Women in Fertility Age

DEFINITION OF TERMS

Placental Malaria: Malaria parasites found in the placenta following maternal infection with *Plasmodium* species during pregnancy.

IPT-SP: Intermittent preventive treatment with Sulfadoxine pyrimethamine is a drug given to pregnant women at regular intervals to clear malaria parasites as well as prevent malaria infection.

IRS: Periodic Spraying of houses with insecticides aimed at killing the malaria vector in other to reduce malaria incidence in the community.

Low Birth Weight: A baby with less than 2.5kg weight at birth.

Malaria Parasite Density: Total number of malaria parasites counting in the blood film per microliter of blood using a microscope.

ABSTRACT

Background

Placental malaria (PM) poses life-threatening complications to pregnant women as they are at increased risk of maternal and neonatal morbidity and mortality. Despite several interventions put in place to reduce malaria in pregnancy in Ghana, latest studies in the northern part revealed one out of every two pregnant women delivering in a hospital have PM. The study determined the proportion of women with PM in the Upper West Regional Hospital (UWR) and its associated factors.

Methods

A cross-sectional hospital-based design with a quantitative approach was carried out among pregnant women delivering at UWR Hospital. The eligible mothers (300) were consecutively recruited and a structured questionnaire used to collect data from the mothers. Placental blood samples were taken for microscopy to determine PM parasitemia. Microsoft Excel 2016 and Stata version 15 were used to analyze the data. Univariate and multivariate analysis were done for the factors using simple and multiple logistic regression respectively. Significance was considered for all variables at $p<0.05$ and 95% confidence level.

Results

The proportion of mothers with placental malaria was 7% (21/300), (95%CI, 4.3-10.5%). Majority of the women (66.7%) with placental malaria had parasite density in the range 501 to 5,000 parasites/ul. Predictors of PM were ANC and gravidity. Whereas primigravida was associated with an increased odd of three times compared with multigravida (aOR=3.48, 95%CI= 1.01 – 12.01), that of less than 4 ANC attendance was associated with nine times increase in odds compared with mothers who attended more than four ANC, (aOR=9.78, 95%CI= 2.89 – 33.11).

Conclusion

The proportion of women with PM was low. Primigravid mothers and less than four ANC visits were associated with PM.

All pregnant women especially primigravida should be encouraged to attend ANC of at least four visits before delivery.

CHAPTER ONE

INTRODUCTION

1.1 Background

Malaria in pregnancy is a serious public health issue globally as it poses life-threatening complications to both the mother and the fetus (Okell, Griffin, & Roper, 2017). The severity and vast majority of pregnant women with malaria are found in Sub-Saharan Africa where over 25 million cases are recorded annually (Ndeserua, Juma, Mosha, & Chilongola, 2015). Malaria is endemic in most African countries hence majority of women who get pregnant get infected with the disease. This implies that women resident in Africa are at an increased risk to malaria when they get pregnant. Those women who become symptomatic for malaria seek treatment early but a vast majority of the asymptomatic patients are only seen when they develop complications (Bassey, Nyengidiki, & John, 2015). Maternal outcomes varies from anemia, hypoglycemia, sepsis and cerebral malaria (Sharma & Shukla, 2017). Maternal anemia is as a result of infected red cells being hemolyzed. Pregnancy is a state that alter the general immunity of the mother and so infection with malaria further lowers the immune system and makes them susceptible to other infections that may lead to sepsis. Fetal complications result from the placenta being infected with malaria parasites and hence leads to spontaneous miscarriages, intrauterine growth restriction(IUGR), low birth weight (LBW), preterm delivery, prematurity, still births and congenital malaria (Sharma & Shukla, 2017).

Placental malaria in sub-Saharan Africa is usually from infection with *Plasmodium falciparum* species (Griffin et al., 2012). Following infection with *Plasmodium falciparum* in the mother's erythrocytes, the parasites are transported to the placenta where they adhere to the intervillous spaces of the mother's site of the placenta thereby resulting in its complications to the fetus and

the mother. Among the many documented factors that contribute to a high risk of placental malaria with *P. falciparum*, nulliparous women stand a greater risk compared with multiparous women. This is because infection in pregnancy leads to expression of *P. falciparum* erythrocyte membrane binding protein (PFEMP1) known as Variant2 chondroitin sulfate antigen (VAR2CSA) peculiar to pregnant women only. The expression of this antigen enables parasitized red blood cells (RBC) bind to oncofetal chondroitin sulfate receptor in the placenta thereby resulting in sequestration of the parasites in the placenta (Ofori, Lamptey, Dickson, Kyei-Baafour, & Hviid, 2018). Immunity to VAR2CSA is not acquired until subsequent pregnancy hence primips are more at risk to placental malaria compared with multips.

The proportion of women with placental malaria in Africa varies depending on endemicity as well as adherence to various interventions that are in place to control malaria among pregnant mothers. Many countries in Africa have National guidelines for control of malaria which is spearheaded by their national malaria control programme (NMCP) department. In Tanzania, a hospital based cross-sectional study revealed 8% prevalence of placental malaria (Ndeserua et al., 2015). Some regions have recorded as high as 65.2% prevalence of placental malaria (Bassey et al., 2015). There are currently several interventions aimed at controlling malaria in all groups with some specific to malaria in pregnancy. The World Health Organization (WHO) recommended the use of Sulphadoxine pyrimethamine (SP) as intermittent preventive treatment (IPTp) of malaria for all pregnant women in malaria endemic areas. The initial 3 doses have been increased to at least 5 doses starting from quickening (16 weeks gestation) to delivery, administered at monthly intervals. Ghana has since adopted this but coverage of the 5 doses still remains an issue in its early days.

1.2 Problem Statement

Globally, over 50 million pregnant women are at risk of malaria in pregnancy annually (Boateng & Anto, 2017). In Ghana malaria in pregnancy accounts for 14% of Out Patients Department (OPD) attendance, 11% of hospitalized admission and 9% of maternal deaths (Wanjiku, 2011). Despite several interventions put in place to reduce malaria in pregnancy and placental malaria in Ghana, the prevalence of placenta malaria is still high. It ranges from 35.9% in rural areas in the southern sector (Ofori et al., 2009) to 52% in the northern sector (van Spronsen, Schneider, & Atasige, 2012).

If the factors contributing to the occurrence of placental malaria are not known, placental malaria will continue to pose life-threatening complications such as IUGR, LBW, congenital malaria, spontaneous abortions, still births, preterm delivery, prematurity and increased neonatal mortality (Singh, Soni, Mishra, Singh, & Bijesh, 2014). The mother will develop anemia, increased risk of postpartum hemorrhage, puerperal sepsis and to a lesser extent hypoglycemic episode in pregnancy as a result of consequence of placental malaria

Adherence to interventions such as insecticide treated net (ITN) use during pregnancy, intake of more than 3 doses of IPTp-SP and appropriate and early treatment of malaria during the first trimester are interventions to reduce malaria in pregnancy and placental malaria. Ghana has since adopted an increase to 5 doses of IPTp-SP for malaria prevention in pregnancy. It is hopeful that this increase, coupled with other malaria prevention interventions in pregnancy, will see significant reduction in placental malaria prevalence especially in the northern zone.

However, latest studies revealed one out of every two pregnancies with placental malaria in northern Ghana (van Spronsen et al., 2012). This study assessed the factors associated with placental malaria in Upper West Regional Hospital.

1.3 Conceptual framework

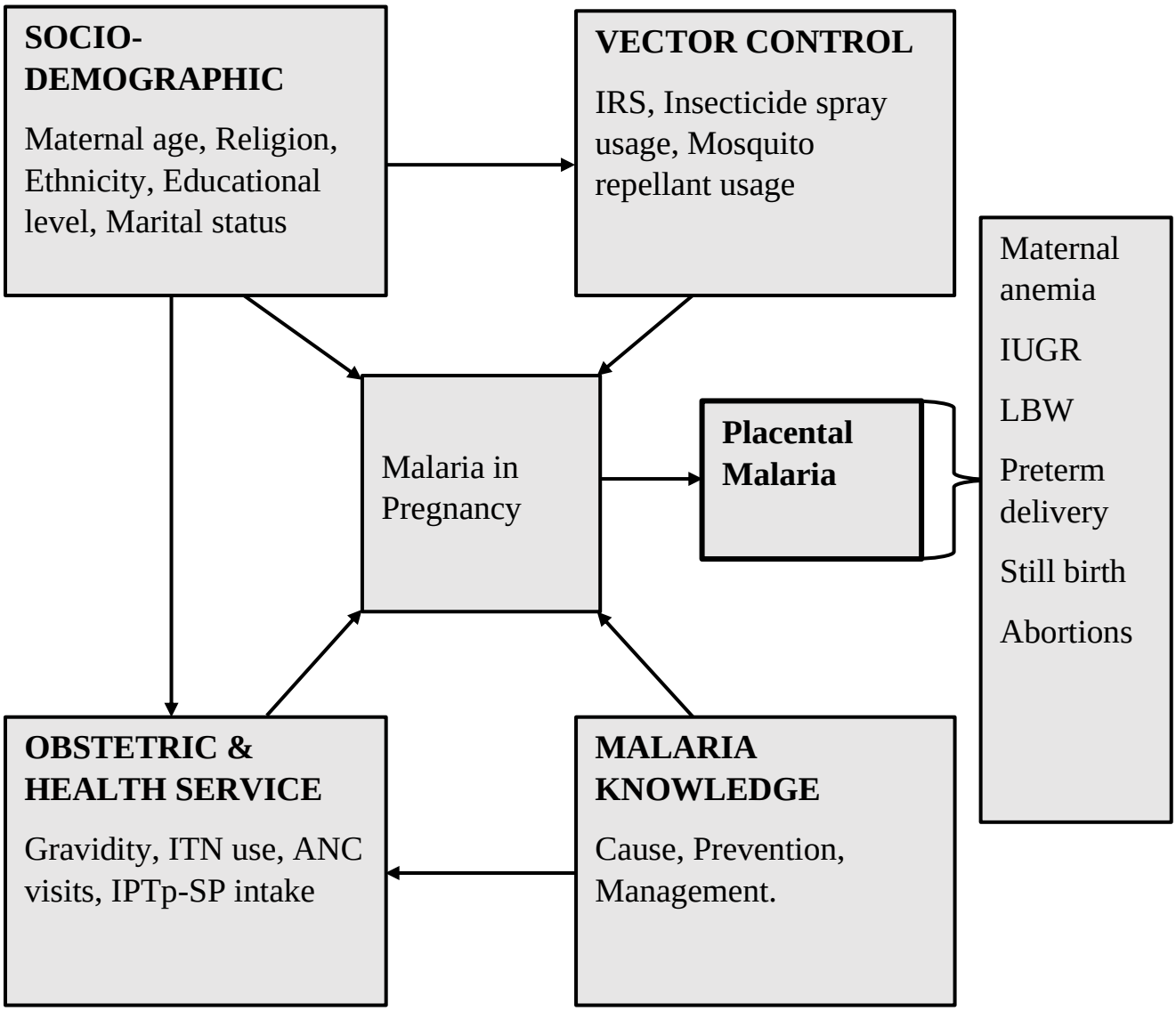

Figure 1: Conceptual framework of factors associated with placental malaria

1.4 Narrative Summary of Conceptual Framework

Malaria in pregnancy is directly linked to placental malaria as *P. falciparum* infection during pregnancy results in sequestration of parasites in the placenta which affects both the mother and the developing fetus. The factors associated with malaria in pregnancy have been grouped into socio-demographic, obstetric and health service factors, knowledge level of the mother and vector

control factors as seen in figure 1. The number of times the mother has been pregnant (Gravidity) will determine exposure to antibodies of malaria parasite and so more pregnancies may lead to low risk of PM and ultimately fewer or no complications.

Maternal age and educational status of a mother will influence how many children a mother would have and also the relative ease at which she will accept interventions such as ITN use and SP intake. This will ultimately influence the risk of infection with malaria during pregnancy and hence placental malaria with its attendant unfavorable pregnancy outcomes. Other vector control mechanisms like Indoor Residual Spraying (IRS), use of mosquito repellants and wearing protective cloths will limit exposure to the vector and hence reduce malaria infection.

1.5 Justification

The increase to 5 doses of IPTp-SP coupled with increase in ITN distribution to pregnant women is supposed to reduce if not eliminate placental malaria and indirectly lead to favorable pregnancy outcomes. However, prevalence of 52% placental malaria in the northern part of Ghana is still of concern although there has been reduction in prevalence by 43% between 2000 and 2006 in southern part of Ghana (Hommerich et al., 2007). Similarly Brong Ahafo region has seen a reduction from 86% to 37% from 2001 (Cox et al., 2005) This study will reveal the proportion of women with placental malaria and the associated factors at the Upper West Regional Hospital. This will serve as baseline data for subsequent studies as well as provide priority areas for malaria control programs to concentrate on for malaria prevention in pregnancy. Also, if the factors associated with PM are known, appropriate interventions can be put in place to address these factors thereby reducing the life-threatening complications that PM poses to the mother and the fetus.

1.6 Study Objectives

1.6.1 General Objective

This study determined the factors associated with placental malaria in the Upper West Regional Hospital.

1.6.2 Specific Objectives

1. To determine proportion of mothers with placental malaria.
2. To assess socio-demographic factors associated with placental malaria.
3. To assess obstetric and health service factors associated with placental malaria.

CHAPTER TWO

LITERATURE REVIEW

2.1 Global Burden of Malaria in Pregnancy

Malaria infection in pregnant women is a public health issue because of the greater number of pregnant women at risk and also the life-threatening complication to this vulnerable group. Worldwide, nearly 244 million women who become pregnant suffer from malaria each year (Wilson et al., 2011). Majority of the cases are from malaria endemic countries. According to Schantz-Dunn & Nour (2009), nearly 50 million pregnant women in malaria endemic areas in the world suffer from malaria every year. Over a million of these cases are co-infected with HIV.

Sub-Saharan African women are among the most burdened with malaria during pregnancy mainly because of its endemicity, accounting for over 25 million cases each year (Fehintola et al., 2016). A direct consequence of this high burden is maternal mortality attributed to the infection. Over 10,000 pregnant women die annually from malaria related infections and nearly 200,000 babies born to malaria infected mothers die before day 28 of life every year (Schantz-Dunn & Nour, 2009).

2.2 Placental malaria burden

Placental malaria results from sequestration of *Plasmodium* species in the placenta following maternal malaria infection. Over 90% of placental malaria results from *P. falciparum* infection, which is the most common parasite causing malaria in endemic countries (Ayres Pereira et al., 2016). India is one of the most populated countries in the world and unluckily a place where malaria is endemic. A tertiary hospital based prospective observational study in Rewa, India

showed placental malaria of 11.8% using blood film microscopy (Singh et al., 2014). The species contributing to this were *P. falciparum* and *P. vivax*.

The prevalence of placental malaria is high in Africa despite interventions aimed at preventing malaria in pregnancy (MiP). A cross-sectional hospital base study in a teaching hospital in southern Nigeria conducted during the rainy season reported placental prevalence as high as 69.3% on placental histology diagnosis and 63.3% on peripheral film blood smear microscopy (Ezebialu et al., 2012). Similar cross-sectional studies in northeastern part of Nigeria conducted during the period of the year where there was no rain yielded a little below 50% placental parasitemia via histology (Bako et al., 2009). The high placental malaria prevalence partly depends on the seasonality in malaria endemic countries. Another hospital based cross-sectional study in Nigeria recorded placental prevalence of 48% (Fehintola et al., 2016). In Tanzania the proportion of mothers with placental malaria could be as high as 75.5% (Menendez et al., 2000) to as low as 8% (Ndeserua et al., 2015).

Ghana shares border with neighboring Burkina Faso to the north and the weather conditions are somewhat similar to the Upper West Region of Ghana. The Placental malaria prevalence in Burkina Faso is as low as 9.1% in rural areas and 4.1% in urban areas (Cisse et al., 2016). A study done in sub-urban coastal region of Ghana documented placental prevalence of 2.5% via blood film microscopy (Stephens et al., 2017). A study done in rural community of Accra along the coast of Ghana recorded placental prevalence of 35.9% (Ofori et al., 2009).

In a prospective study in Brong Ahafo region of Ghana where parasitemia levels are high during the raining season, the study reported a placental malaria prevalence of 38% (Hommerich et al., 2007) via placental histology. Similar appreciable high prevalence of placental malaria (35%) have been recorded in Agogo, southern part of Ghana. This prevalence was recorded in the early 2000

when three doses of IPTp-SP was the recommended malaria prevention prophylaxis for pregnant women by WHO (Mockenhaupt et al., 2006). Decline in placental malaria have been recorded in recent times in southern Ghana where prevalence has hit as low as 15% (Hommerich et al., 2007).

2.3 Factors associated with Placental Malaria

Many factors have been documented in literature to be associated with placental malaria. A number of these factors are prevalent in certain geographic areas where malaria is endemic and significantly correlates with placental parasitemia.

2.3.1 Socio-demographic factors

The age of a woman when pregnant poses a risk of developing placental malaria. Mothers who are younger and entering pregnancy is known to have increased risk of developing placental malaria compared with older age groups. A study done by Babalola, Idowu, Wobo, & Fabusoro, (2015) showed significant association between age range 18 to 22 years and placental malaria. In that study, mothers who were in the age range of 18 to 22 years were four times at risk of developing placental malaria compared to older women. Another study conducted in Cameroon showed a two times increased in odds of developing placental malaria among women 20 years or less compared with those older than 30 years (Tako et al., 2005). Similarly, a study in Sudan by Elhassan et al., (2017) showed a three times increased odds among women aged less than 24 years and this was statistically significant. Other research have demonstrated no relationship between maternal age and placental malaria (Ezebialu et al., 2012; Fehintola et al., 2016; van Spronsen et al., 2012).

The marital status of women is an important factor as male partners are now being actively involved in the care of the woman during antenatal care. It is known that when their partners are

involved in the health care process, the women are more likely to utilize health services offered them. A study conducted in Ethiopia to this effect showed an increase in odds among women (with male partner involvement) attending their first antenatal care in the first trimester and also complying with the number of ANC visits recommended by the health care provider before delivery compared to those that were not married (Mohammed, Johnston, Vackova, Hassen, & Yi, 2019). Most studies have however not demonstrated significant association between marital status and placental malaria (Bako et al., 2009; Cisse et al., 2016; Fehintola et al., 2016; Ibanga et al., 2015).

For occupational status, most studies have demonstrated no association with placental malaria. Uneke, (2008), reported no association between the mother's occupation and placental malaria. In Africa, most women leaving in rural areas assist their husbands on their farms and some are independently farmers. Although these women who are farmers should be at a greater risk to mosquito bites, studies have not reported any association between placental malaria and farmers.

Maternal education cannot be underestimated in terms of its correlation with maternal and child health. Most studies have demonstrated good pregnancy outcomes among women with higher educational level (Karlsen et al., 2011). In terms of placental malaria, Elhassan et al., (2017) showed a two times increase in odds among mothers who were not educated at the secondary school level compared with their counterparts who had at least secondary level education.

A study in Nigeria showed that being resident in a rural areas community increases the mothers chances of developing placental malaria compared with their urban counterparts (Ezebialu et al., 2012). According to Babalola et al., (2015) pregnant mothers sleeping in a congested room (more than 3 occupants) have 60% increase risk of placental malaria. When there are more occupants in

a sleeping area, there will be frequent opening of the doors to the sleeping area and as such making them prone to bites of the female anopheles' mosquitos.

2.3.2 Obstetric and health service factors

Mothers who are pregnant for the first time (nulliparous women) stands a higher risk of placental malaria compared to their multiparous counterparts (Asante et al., 2013; Ezebialu et al., 2012; van Spronsen et al., 2012). A study among Sudanese women showed a threefold increase in placental malaria among primigravid mothers compared to mothers in their second or more pregnancy (Elhassan et al., 2017). Similarly, Bouyou-akotet et al., (2003) also reported an increase association among Gabonese nulliparous women.

Despite an increasing concern of IPT-SP resistance (Okell et al., 2017), most studies have reported protection against placental malaria with intake of at least 3 doses of IPT-SP. It has been documented that women receiving at least three doses of SP reduces plasmodium sequestration in the placenta (Anchang-Kimbi et al., 2014; Ayres Pereira et al., 2016; Wilson et al., 2011). However some other studies have demonstrated no association between the number of IPTp-SP doses and placental malaria (Elhassan et al., 2017; Ndeserua et al., 2015; van Spronsen et al., 2012).

Mothers who own and sleep in ITN during pregnancy have a reduce risk of developing placental malaria compared with those who don't sleep in it. A study among Nigerian pregnant mothers showed a two times increased risk of PM among women who did not sleep in ITN prior to delivery (Babalola et al., 2015).

Co-infection has been shown to increase the risk of placental malaria, especially among HIV pregnant women. Pregnancy is a state where the immune system is lowered and so pregnant women are susceptible to most febrile infections other than malaria. A study done in Benin found

significant association between non-malaria febrile episodes such as gastrointestinal disorders and respiratory tract infections and placental malaria (Rachas et al., 2012).

During ANC visits mothers are educated on the state of their pregnancy, likely complications they may encounter and how to prevent and manage infections including malaria. It's been shown that mothers who regularly attend ANC has a reduce risk developing malaria and hence placental malaria compared to those who do not go for ANC (Anchang-Kimbi et al., 2014). Similarly Elhassan et al., (2017) in their study among Sudanese woman reported an eleven times increase odds of having placental malaria in those women who did not attend antenatal care prior to delivery. It is therefore evident that attending the recommended ANC visits prior to delivery will result in good pregnancy and delivery outcome.

2.4 Malaria prevention and control during pregnancy

There are a number of measures to actively practice in order to prevent malaria infections. Most countries have implemented and are practicing the WHO recommendations for preventing and controlling malaria in pregnancy. WHO recommends every pregnant woman to sleep in ITN throughout the pregnancy period, intermittent chemoprophylaxis with Sulphadoxine-pyrimethamine (SP) and early diagnosis leading to effective treatment of malaria as the three main interventions to control malaria in pregnancy. The benefits of IPTp-SP intake include not only prevention of MiP but also prevention of mothers developing anemia, placental malaria and improve new born weight as well as reduction in neonatal mortality (WHO, 2012).

The initial recommended 3 doses of IPTp-SP was implemented in Ghana in 2005 (Nwaefuna, Afoakwah, Orish, Egyir-yawson, & Boampong, 2015). Although coverage has been an issue, the

effectiveness of SP in preventing and controlling malaria has been showed in a number of studies (WHO, 2012). An evaluation studies by Hommerich et al., (2007) showed over 43% significant reduction in *P.falciparum* among women on SP. The number of SP doses has been increased to five doses as recommended by WHO following numerous evidence on its effectiveness in many studies.

Sulphadoxine Pyrimethamine is given as a single dose medication starting from 16 weeks gestation. It is a directly observed treatment drug and so the issue with compliance is not a problem since the healthcare worker directly observes the pregnant woman taking it. The current recommended five doses of SP was adopted in Ghana as a policy in 2014 and like the previous three doses, coverage is an issue in its early years of implementation. Administration of the SP starts at 16 weeks of gestation and given at monthly intervals during antenatal visits until delivery. To be able to get all the five doses means the mother must start ANC early (at 16 weeks) and attend at least 5 times at monthly intervals. A number of factors influence ANC visits and hence coverage of IPTp-SP, notable among these are: access to health facility providing such services, maternal educational level, socio-economic status, attitude of health workers providing ANC services and religious as well as cultural beliefs (Nsibu et al., 2016). Contraindications to SP intake during pregnancy include, women receiving cotrimoxazole and Glucose 6 Phosphate Dehydrogenase Deficiency (G6PD) patients (WHO, 2012). In addition, folic acid of 5mg or more should not be administered concomitantly with SP because of its counteraction which reduces therapeutic efficacy for malaria treatment.

There has been increasing concerns of SP resistance with enormous studies conducted to determine molecular markers prevalence such as dihydrofolate reductase and dihydrofolate synthase (dhfr/dhps) and resistance to SP. A research conducted in Tanzania showed significant association

between parasitemia levels of malaria in the placenta and resistant mutant allele at dhps codon 581 (Harrington, Mutabingwa, Kabyemela, Fried, & Duffy, 2011; WHO, 2012).

Another malaria intervention which is fast gaining promise in malaria endemic countries is the vector control mechanism using chemicals to spray homes (indoor residual spraying-IRS). Although acceptability has been the concern in its early days, studies in areas where it has been implemented have shown reduction in the prevalence of malaria cases. In Benin a study recorded a decrease by 8% the incidence rate compared to 18% in areas that were not exposed to IRS (Ogouyèmi-hounto et al., 2018). This intervention offers protection in pregnancy as well. A study conducted in Uganda among pregnant women found a 16.8% higher prevalence of placental malaria (P=0.001) in those who had no IRS protection compared with women who had IRS protection (Muhindo et al., 2016).

2.5 Diagnosis of Placental Malaria

Laboratory confirmation of plasmodium species in the placenta following sequestration is mainly by 4 methods, blood film microscopy, immunochromatography to detect to detect Histidine Rich Protein 2 (HRP2) via rapid diagnostic test (RDT) kits, polymerase chain reaction (PCR) on placental tissue and histopathology. The sensitivity and specificity of all these test are different with histopathology being the gold standard (Campos et al., 2011). Histopathology and PCR yields high specificity but have the limitation of being costly and cumbersome requiring expertise to carry out the test and hence limits field use in malaria endemic regions.

Light Microscopy diagnosis involves taking blood sample usually about 2mls from the intervillous space of the maternal site of the placenta and preparing thick and thin films stains in order to

visualize the malaria parasite species types as well as quantify the parasites using the light microscope. Light microscopy diagnosis of malaria is influence by: technical expertise of the reader in terms of experience, how the sample is prepared and the number of parasites in the sample (Graffeo et al., 2008). Parasitemia levels more than 1,000/L yields better results on light microscopy (Campos et al., 2011). For RDT, the blood sample from the placenta is tittered onto the test kit and the results read as positive, negative or indeterminate.

To diagnose placental malaria via histopathology, placental tissues are prepared and stained with Hematoxylin – Eosin (H-E) and the slides read by a pathologist. Histopathology can classify placental malaria into acute, chronic, past and uninfected placenta.

For PCR diagnosis the placental tissue is taken and PCR assayed for presence or absence of malaria parasite. Many studies have demonstrated discordance in placental malaria parasitemia with RDT, light microscopy, PCR and histopathology. One such study is the Columbia study which showed a 17% discordance in placental malaria parasite among the study group for microscopy (10%) and PCR (27%) (Campos et al., 2011). The sensitivity and specificity of placental blood microscopy compared to the gold standard of histopathology is 76% and 99% respectively (Mockenhaupt et al., 2006). Similar studies in Ghana showed prevalence of placental malaria as 59.4%, 40.8% and 30.9% for PCR, RDT and microscopy respectively.

2.6 Treatment of malaria in pregnancy

There are several drugs recommended for treating malaria but few of these are safe in pregnancy. The first trimester of pregnancy is a very critical point for organogenesis and so not all the known

antimalaria drugs are recommended for use. Malaria infection can either be uncomplicated or severe and so what medication to use depends on this.

The World Health Organization (WHO) recommends the use of quinine plus clindamycin for first trimester treatment of simple malaria in pregnancy as first line (WHO, 2007). When clindamycin is not readily available, monotherapy with quinine alone can be used. Artemisinin-based combination therapies (ACTs) such as artemether lumefantrine can be used only when the first line drug is not available.

There has been varying concerns about the safety of ACT in the first trimester with little evidence on its harmful effect (Pekyi et al., 2016). For second and third trimester pregnancy, ACT is safe and is the drug of choice by WHO for treating simple malaria. First line antimalaria used to treat simple malaria for pregnant women who are 13 weeks onwards is ACT or artesunate plus clindamycin or quinine oral.

Severe malaria mostly requires admission and the WHO recommend parenteral antimalaria. Intravenous (IV) quinine or IV artesunate is the drug of choice for treating severe malaria. Quinine has the side effect of hypoglycemia and so its administered with intravenous dextrose.

The most concerned harmful effects with the use of ACT in the first trimester treatment of malaria is miscarriage and congenital abnormalities. Some studies have documented no difference between quinine and ACT in causing miscarriage or congenital abnormalities (Moore et al., 2016; Pekyi et al., 2016).

2.7 Antenatal Care

Antenatal Care (ANC) according to WHO (2016) is "*the care provided by skilled health-care professionals to pregnant women and adolescent girls in order to ensure the best health conditions for both mother and baby during pregnancy*". Many maternal deaths arising from complications due to pregnancies could be avoided through regular ANC visits by the pregnant women (Tuladhar & Dhakal, 2012). However ANC attendance is still an issue in most developing countries especially in rural areas (Ruragiriwa, Mogren, Nyirazinyoye, Ntaganira, & Krantz, 2017). The World Health Organization recommends at least 4 ANC visits for low risk pregnant mothers but for high to moderate risk pregnant women as many times as the health care provider suggest.

A study conducted in Kumasi, Ghana, showed 10% of pregnant women did not meet the recommended ANC attendance (Asundep et al., 2013). Notable reasons for not attending ANC by these pregnant women included, not wanting to know their retro status, cost, accessibility, having drugs at home and laziness among others. In Rwanda the proportion of women who did not meet the recommended ANC attendance was five times higher (54%) (Ruragiriwa et al., 2017) than what the Ghana study found. In the Rwanda study, inadequate or no ANC visit was significantly associated with older women, single mothers and less affluent women. Another study in Ghana reported 74.6% of pregnant women meeting the recommended ANC visits (Fondjo et al., 2018).

During ANC, pregnant women are provided with education on their pregnancy spanning from signs and symptoms of complications and unfavorable state, nutritional support and advice, provision of services such as immunization and essential drug supplements. An important part of ANC in relation to placental malaria is provision of IPT-SP to the women as well as early diagnosis and treatment of malaria. This visit is also an opportunity for the health care provider to offer insecticide bed net to the mother in Ghana.

2.8 Consequence of placental malaria

Sequestration of malaria parasite in the developed placenta poses deleterious effects to both the mother and the developing fetus. Maternal effects such as anemia, spontaneous miscarriages and maternal deaths have been reported following placental malaria infection (Uneke, 2007).

In sub-Saharan Africa nearly half a million pregnant women suffer from malaria related anemia and about 10,000 of these women die from it every year (Uneke, 2008). A hospital based cross-sectional study in Tanzania reported a twofold increase in odds of anemia among women with placental malaria (Ndeserua, Juma, Mosha, & Chilongola, 2015). Another study in Cameroun reported statistically significant association between PM and maternal anemia (Tako et al., 2005).

Adverse effects of placental malaria to the fetus include IUGR, preterm delivery, prematurity, congenital malaria, low weight babies and fetal anemia. Many studies have demonstrated significant association between placental malaria and babies born with weight less than 2kg. A study in Nigeria showed 32% of new borns with weight less than 2kg born to mothers with placental malaria compared to mothers with no placental malaria (Oraneli, Okeke, & Ubachukwu, 2013).

CHAPTER THREE

METHODS

3.1 Study design

The study was a cross-sectional study with a quantitative approach conducted in the Upper West Regional Hospital Maternity Unit. This study involved pregnant women who presented at the labor unit of the regional hospital for delivery. The mothers were interviewed using structured questionnaire and their ANC book reviewed with a checklist. There was a laboratory component where placental blood was taken for microscopy. The data from the questionnaire was analyzed descriptively and expressed as frequencies, means and standard deviations. Logistic regressions were used to determine association between dependent and independent variables using Stata software.

3.2 Study Area

The Upper West Region of Ghana was the location for the study and conducted specifically in the regional hospital at the Maternity unit and labor ward of the hospital. The regional hospital is the major referral center of the region, serving inhabitants of the region and neighboring Burkina-Faso. The hospital is located at the central part of the regional capital-Wa and so its surrounding environ is urban.

The hospital has a total of 96 beds and a monthly average delivery of 200. The hospital has four departmental units: obstetrics and gynecology unit, medical unit, pediatric unit and surgical unit. The labor ward is in the obstetrics and gynecology unit. Deliveries are high during the last quarter of the year but low in the first quarter of the year (hospital records).

The region is located in the north-western part of Ghana, lies between longitude 1° 25'' W and 2° 45'W and latitudes 9° 30'' N and 11°N. To the south of the region is the Northern region, to the north and west is Burkina Faso and to the east is Upper East region.

The Upper West region has a tropical climate and so temperatures can be as low as 22.6°C and as high as 40.0°C during the year. Unlike the southern part of Ghana, this region has a single rainy season usually from May- October. The rain intensity is often between 100-115 cm/annum and humidity ranging between 70% - 90% but falling to 20% in the dry season.

There are three hundred and thirty-three (333) health facilities within the region. These include eleven (11) hospitals (government, private and CHAG). The rest are four (4) Polyclinics, seventy (70) health centres, fifteen (15) clinics, two hundred and twenty-seven (227) CHPS Compounds and five (5) maternity homes.

The total population of the region is 786,050 of which women in the fertile age (WIFA) forms 24%. Figure 2 shows the map of Ghana showing location of the Upper West Regional hospital in the Upper West region.

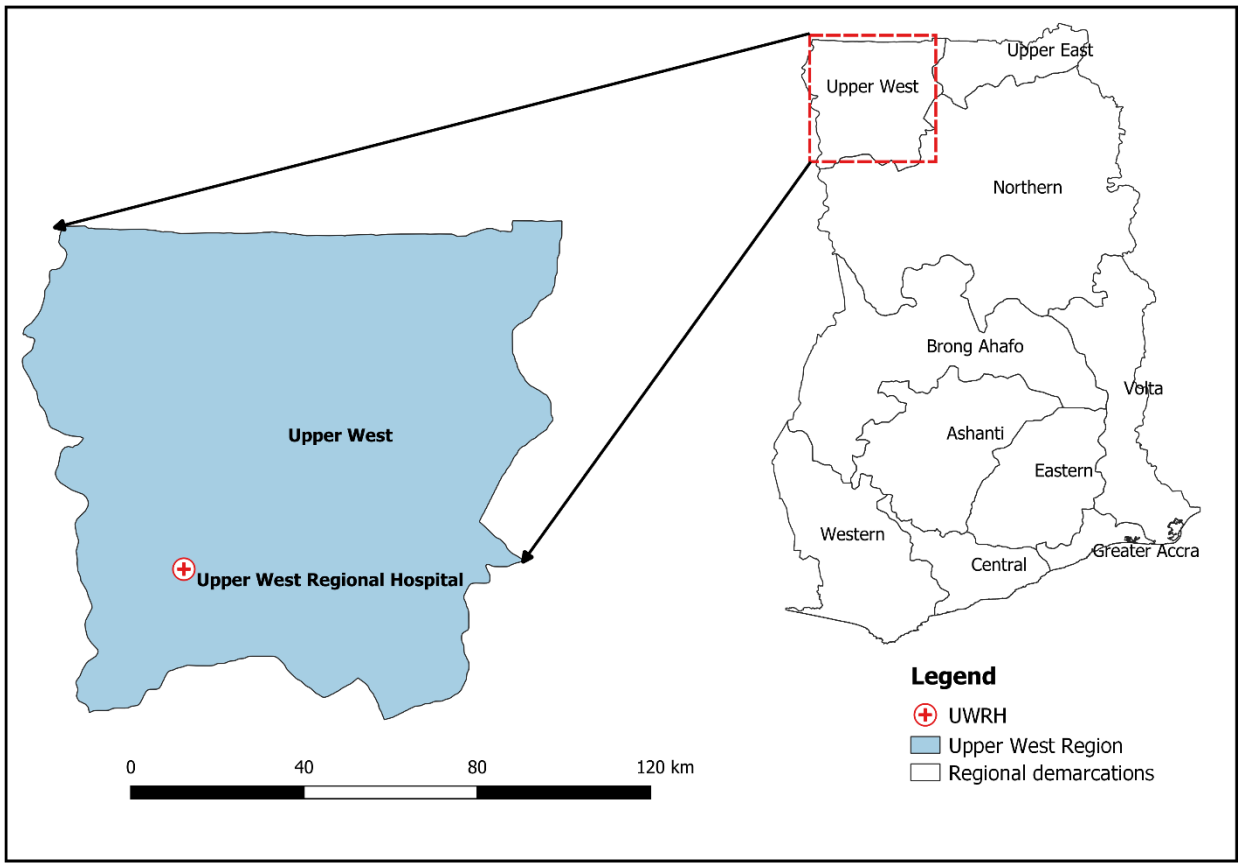

Figure 2:Map of Ghana showing Wa Regional Hospital located in Upper West Region

3.3 Study Variables

The outcome variable for the study was placental malaria. Placenta malaria was assessed as positive following microscopy results of placental sample showing malaria parasites via Giemsa staining. Negative samples were those that did not show malaria parasites following microscopy via Giemsa staining.

Independent variables were maternal age, educational status, marital status, religion and ethnicity. Others such as Gravidity, ITN use, SP intake and number of ANC attendants were also assessed as independent variables. Table 1 shows the variables measured.

Table 1: Variables measured

Variable	Operational definition	Scale of Measurement
Dependent variable		
Placental malaria	Plasmodium parasites seen in placental blood sample via microscopy	Categorical
Independent variables		
Socio-demographic factors		
Age of mother	Age at last birthday	Continuous
Ethnicity	Ethnic background of mother	Categorical
Religion	Type of religion practiced by mother	Categorical
Occupation	Occupation of mother	Categorical
Level of education	Highest level of education attained by mother	Categorical
Marital status	Marital status of mother	Categorical
Obstetric & health Service		
Gravidity	Number of times mother has been pregnant	Categorical
Use of ITN	Mother sleeping under ITN during pregnancy or within the last 7 days	Categorical
SP intake	Number of SP doses taken during pregnancy	Continuous
ANC visit	Number of ANC visits by mother during pregnancy	Categorical
IRS	IRS at mother's house	Categorical
Hypertension	Systolic blood pressure greater than 130mmHg and diastolic ≥90mmHg	Categorical
Maternal Hb	Hb of mother at 36 weeks	Continuous
Malaria in index pregnancy	Diagnosed with malaria or received treatment for malaria from week 1 to term pregnancy	Categorical

3.4 Study Population

The study respondents were pregnant women who delivered at the Upper West Regional Hospital and met the inclusion criteria for the study.

3.5 Inclusion and Exclusion Criteria

3.5.1 Inclusion criteria

All pregnant women who delivered at the Upper West (Wa) Regional Hospital and consented to the study were included.

3.5.2 Exclusion criteria

Pregnant women who were Glucose 6 phosphate dehydrogenase positive, those who's pregnancy are less than 28 weeks at the time of delivery and all pregnant women with serious medical conditions such as renal failure were excluded.

3.6 Sampling

3.6.1 Sample Size

The total sample size used was 300.

The sample size was determined using the following parameters derived from Cochran's formula below

$n=z^2*(pq)/d^2$

n= sample size

z= critical value at 95% confidence level which is 1.96

p= prevalence of placental malaria which is 15% (0.15) (Hommerich et al., 2007)

q= 1-p which is 0.85

d= permitted error of margin which is 5% (0.05)

The minimum sample size calculated was 196

Considering removal of incomplete responses as well as increasing the power of the study, a total of 300 women was used.

3.6.2 Sampling technique

Pregnant women admitted for delivery at the labor ward of the Regional Hospital who were eligible per the inclusion criteria were consecutively recruited as study participants to make up the sample size of 300 from January 2019 to March 2019. Every eligible mother who presented for delivery was selected until the 300th person was recruited within the three-month period.

3.7 Data collection techniques and tools

A structured questionnaire and checklist were designed using the software Enketo to collect maternal data from their antenatal health record book. The mothers were interviewed after delivery with structured questionnaire pertaining to questions on their socio-demographics, the use of insecticide treated bed nets and IRS exposure among others. The checklist was used to collect maternal data from their ANC record book and this included their obstetric variables, medical history and health facility factors. The maternal records data were cross checked by asking the mothers same questions. For the new born, their weight and outcome such as still birth and preterm were collected from the deliveries record card.

Trained research assistants (Midwives) helped in collecting the data using android mobile devices that had the downloaded Enketo software questionnaire and checklist.

Eligible participants had their placental blood taken immediately after delivery of the placenta. The sample taking was carefully done and due processes and protocols followed. A trained laboratory technician was at hand to take the samples.

3.8 Placenta blood sample taking process

The laboratory technician was appropriately gowned, gloved and masked. The delivered placenta was placed in a wide kidney dish with the maternal side facing upwards.

A sterile soaked gauze with savlon was used to clean the maternal site of the placenta. An incision of about 1.5cm deep was made in the mother's part of the placenta through the intervillous space with a scarpel blade and a 5cc syringe used to collect 3mls of placental blood from the pool of blood into ethylenediaminetetraacetic acid (EDTA) bottle.

The bottle was labeled with the maternal ID corresponding to the questionnaire.

3.8.1 Transport and processing of placenta blood sample

The blood sample was immediately transported in a carrier box at room temperature to the laboratory within the hospital on the same day for analysis. Thick and thin stains were prepared using 3% Giemsa stain following standard protocol. The slides were read by a biomedical scientist under light microscope and those that were positive for malaria parasites were recorded in terms of parasite density (counts). The species type was also recorded. The laboratory results were then matched with the maternal ID on the questionnaire for entry into the Enketo software.

3.9 Pre-testing of questionnaire

The questionnaire was pre-tested on 10 respondents in Nadowli Hospital located in the upper west region. Nadowli hospital is similar in terms of number of deliveries performed as well as close proximity to the location of the regional hospital.

3.10 Data processing and analysis

Data from the questionnaire on the enketo software was extracted from a central database and then to Microsoft Excel 2016. Data cleaning was done in Microsoft Excel and from there imported into STATA version 15 for analysis.

Descriptive statistics was performed for all variables and expressed as frequencies, means and standard deviations for continuous data as well as medians and interquartile range for variables such as age.

Numerical variables were summarized as percentages and cross-tabulations done for categorical variables to estimate proportions. The proportion of mothers with placental malaria was estimated via proportion of those respondents who tested positive for placental malaria. The placental malaria parasite density was also estimated. The determined malaria parasite density was further categorized into low parasite density, moderate parasite density and high parasite density according to the WHO classification. Low parasite density is malaria parasite density $\leq$ 500 parasites/µl, Moderate parasite density is from 501 parasites/µl to 5,000 parasites/µl and high parasite density is > 5,000 parasites/µl. Those samples that were positive also had the malaria parasite species recorded.

Logistic regression was done to test for association with the outcome variable. Univariate analysis for various factors such as IPTp-SP, maternal age, gravidity and ITN was done to test for associations at the crude level using simple logistic regressions. Crude odd ratios were generated at 95% confidence interval (CI). Multiple logistic regression analysis was also performed to assess the effect of multiple factors on placental malaria using multiple logistic regressions. Factors which had statistical significance at the univariate level were all included in the multivariate analysis to generate adjusted odds ratio. Significance was considered at $p<0.05$ with CI 95%.

3.11 Quality control

Experienced midwives and laboratory technicians were extensively trained on the data collection process as well as the use of the software questionnaire. The trained research assistants were deployed for the pre-testing. For blood film microscopy readings, an experienced biomedical scientist from the Regional Hospital was employed to read the slides. Ten positive placental malaria slides and 10 negative slides were randomly selected to a second private laboratory for reading in other to ascertain the right results. All 20 slides corresponded with the previous results.

The stained slides and samples have been preserved for future use and will not be discarded until data published.

3.12 Ethical and Safety Consideration

The Ghana Health Service (GHS) Ethics Review Committee (ERC) approved the study after reviewing the protocol. The approval reference is **GHS-ERC013/12/18**. Permission was sought

from the Upper West Regional Health Directorate through the Regional Director of Health Services and also from the Regional Hospital Medical Director.

A written informed consent was sought from eligible respondents before recruiting them into the study. The process of consent was explained to the participants in terms of the risk involved, benefits and withdrawal process. Participants were made aware that it was entirely voluntary for participation and could withdraw at any point in time without any consequence.

Data collected were kept confidential and only the Principal investigator and academic supervisors have access to the data via password protection. Codes were assigned to each participant to avoid revealing personal information.

CHAPTER FOUR

RESULTS

The findings of the study which involved 300 mothers and their placental blood samples have been summarized in this chapter. Variables that had not been responded to by participants resulting in less than 300 responses (missing values) have been stated.

4.1 Socio-demographic characteristics of respondents

The age of the participants ranged between 18 years to 48 years and the median age was 26 years interquartile range (p25=22, p75=31). Majority of the participants (46% (138/300)) were between 25 to 34 years. Almost all the respondents were married (94% (282/300)). Most (90.7% (272/300)) of the participants were from ethnic backgrounds dagaare/wala/sisala which happens to be the major ethnic groups of the region. Other ethnic backgrounds captured included Hausa, Fulani and Moshi representing 2% (6/300) of the respondents. For educational status, 17% (51/300) had never been to school.

Traders formed majority (35% (105/300)) of the respondent's occupation whereas Islam was the dominant religion among the participants (64% (192/300)). Table 2 illustrates the socio-demographic characteristics of the mothers.

Table 2: Socio- demographic characteristics of study subjects

Characteristic	Frequency (%)N=300	Placenta Malaria	
		YES	NO
Maternal age			
18-24	126 (42.0)	16	110
25-34	138 (46.0)	4	134
35-44	34 (11.3)	1	33
≥45	2 (0.7)	2	0
Marital status			
Single	5 (1.7)	2	3
Married	282 (94.0)	14	268
Divorced	4 (1.3)	0	4
Co-habiting	9 (3.0)	5	4
Ethnicity			
Dagaare/wala/sisala	272 (90.7)	19	253
Frafra/Kasena/builsa	6 (2.0)	2	4
dagomba/gonja/maprusi	13 (4.3)	0	13
Akan	3 (1.0)	0	3
Other	6 (2.0)	0	6
Religion			
Christianity	107 (35.7)	7	100
Islam	192 (64.0)	13	179
Traditional	1 (0.3)	1	0
Educational status			
No education	51 (17)	2	49
Basic Education	181 (60.3)	13	168
Secondary	39 (13)	4	35
Tertiary	29 (9.6)	2	27
Occupation			
Student/unemployed	53 (17.7)	4	49
Farmer/housewife	91 (30.3)	8	83
Business/civil servants	24 (8.0)	1	23
Trader	105 (35.0)	4	101
Other	27 (9.0)	4	23

4.2 Obstetric and health service characteristics of respondents

Women who were pregnant for the first time (primigravida) formed 29% (87/300) of the respondents and the rest were pregnant for the second or more. Majority 50% (150/100) of the women had received three or more doses of the SP as intermittent preventive treatment for malaria in pregnancy. Individually, those who received three doses were the majority 31.3% (94/300). There were some respondents who never received SP throughout the pregnancy, 7% (21/300).

For ITN use, those mothers who responded to sleeping under a treated bed net during the pregnancy were a little over 50%, however ITN ownership was as high as 85% (255/300).

Women who became pregnant for the first time formed 29% (87/300) of the respondents. Despite the policy of all women getting tested for HIV in pregnancy in order to reduce mother to child transmission, some mothers 2.3% (7/300) were not tested. Table 3 shows the obstetrics and health service characteristics of the study participants.

Table 3: Obstetric and health service characteristics of study participants

Characteristic	Frequency (%) N=300
Gravidity	
Primigravida	87 (29)
Multigravida	213 (71)
IPT-SP Use	
No SP dose	21 (7.0)
Single dose	38 (12.67)
Two doses	91 (30.3)
Three doses	94 (31.3)
Four doses	39 (13.0)
Five doses	17 (5.7)
ITN use	
Yes	178 (59.3)
No	122 (40.7)
ANC attendance	
None Attendant	4 (1.3)
1 to 3 attendance	32 (10.7)
4 or more attendance	264 (88)
HIV status	
Negative	279 (93.0)
Positive	14 (4.7)
Not done	7 (2.3)
Hypertension (n=299)	
Hypertension	7 (2.3)
Normotension	292 (97.7)

For antenatal care attendance, majority 88% (264/300) met the recommended four or more attendance by WHO. There was however some proportion of mothers 1.3% (4/300) who did not attend ANC. The mean ANC attendance was 5.3 (SD 1.7). The ANC attendance ranged between 0 and 10 visits.

Table 4: Maternal obstetric characteristics

Characteristic	Frequency (%)
Fever	
Yes	106 (35.3)
No	194 (64.7)
Malaria treatment in index pregnancy	
Yes	120 (40)
No	180 (60)
Maternal Hb-level	
≥11g/dl	12 (4.4)
≤11g/dl	262 (95.6)
Sickling status	
Negative	285 (95)
Positive	5 (1.7)
Not done	10 (3.3)

Table 4 shows other maternal obstetric characteristic. It can be seen from the table that a significant proportion of the respondents 40% (120/300) were diagnosed with malaria and received treatment during the pregnancy. Anemia among pregnant women was high as nearly all 95.6% (262/284) of the women who had their Hb recorded had less than 11g/dl. The lowest Hb was 5.6g/dl and the highest was 13g/dl. The mean maternal Hb was 9.5g/dl (SD 1.4).

4.3 Environmental and other characteristics of respondents

Most 79.3% (238/300) respondents indicated that their homes have been sprayed during the annual on-going indoor residual spraying (IRS). Only 20.7% (62/300) of the respondents however did not receive IRS exercise in their homes during this index pregnancy. Table 5 shows the environmental characteristics of the respondents.

Table 5: Environmental and other characteristics of respondents

Characteristic	Frequency (%) N=300
IRS	
Expose	238 (79.3)
Not expose	62 (20.7)
Bushy surrounding	
Bushy	47 (15.7)
Not bushy	253 (84.3)
Mosquito repellent	
Use	72 (24)
No usage	228 (76)
Insecticide spray	
Use	98 (32.7)
No usage	202 (67.3)
Sleeping room occupancy	
≤ 2 people	138 (46)
>2 people	162 (54)

4.4 Proportion of women with placental malaria

The proportion of women with placental malaria parasites delivering at the Upper West Regional Hospital was 7% (21/300), (95%CI, 4.3-10.5%). All the placental malaria cases were *Plasmodium falciparum* species. The parasite density ranged from 237 parasites/ µl to 70,821 parasites/ µl. The median parasite density was 2,315 interquartile range(p25=1101, p75= 3860 parasites/µl). Figure 3 shows the parasite density of women whose placenta tested positive.

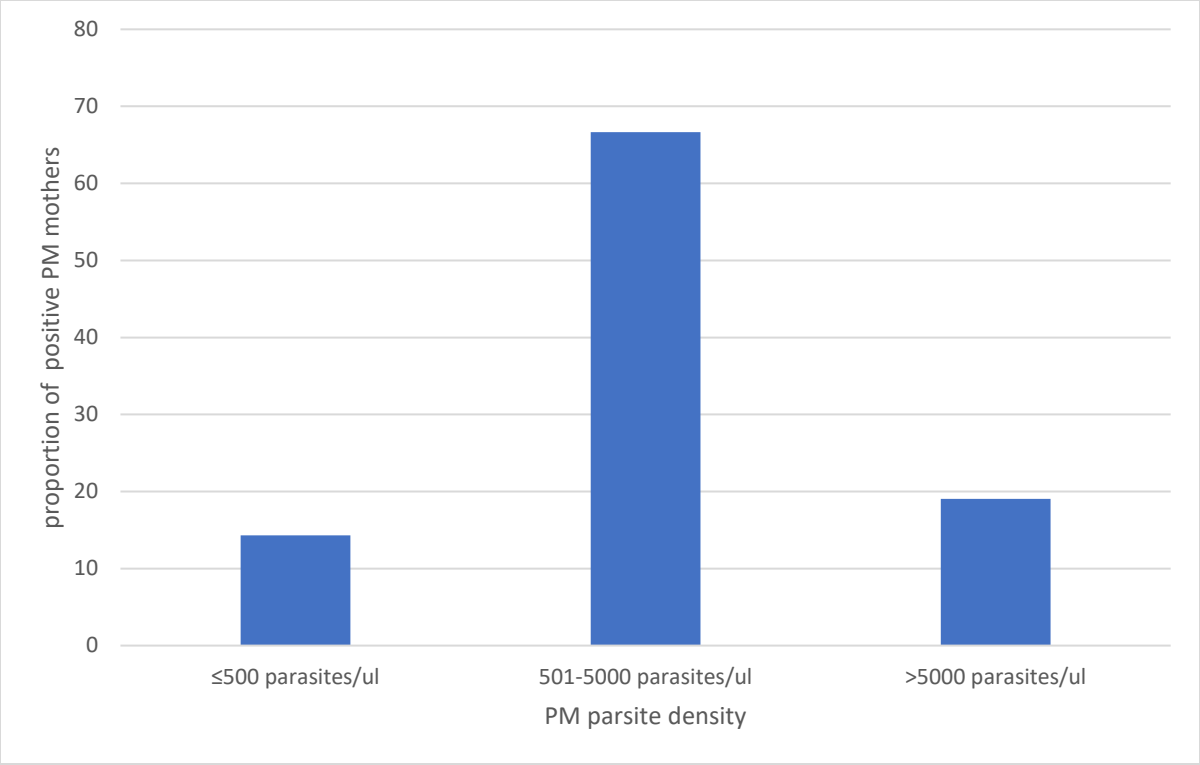

Figure 3: Placental Malaria (PM) parasite density of 21 mothers who tested positive

4.5 Factors associated with placental malaria

This section presents the results of the sociodemographic and obstetric as well as health service factors associated with placental malaria.

4.5.1 Socio-demographic factors

The socio-demographic factors that were significantly associated with placental malaria in Upper West Regional Hospital were maternal age, marital status, and ethnicity. Mothers who were 20 years or less had 6 times increase in odds of having placental malaria compared with mothers aged greater than 20 years (cOR=6.04, 95%CI= 2.42 – 14.09). For marital status, women who were not married had greater odds of placental malaria compared with the married ones (cOR= 12.18, 95%CI= 4.09 – 36.21). Those women who were from the ethnic background Frafra/Kasena/Builsa

had greater odds of having placental malaria compared to the native ethnic group (cOR= 6.66, 95%CI= 1.15 – 38.71). Religion and educational status were not significantly associated with placental malaria. Table 6 shows the univariate analysis of the socio-demographic factors associated with placental malaria.

Table 6: Analysis of socio-demographic factors associated with placental malaria

Characteristics	Placenta malaria		cOR (95%CI)	p-value
	No	Yes		
Maternal age				
≤20 years	43	11	6.04(2.42 – 15.09)	**<0.0001***
>20 years[1]	236	10	1	
Marital status				
Not Married	11	7	12.18(4.09 – 36.21)	**<0.0001***
Married[1]	268	14	1	
Religion				
Islam	179	13	1.04(0.40 – 2.69)	0.939
Christian[1]	100	7	1	
Ethnicity				
Frafra/Kasena/builsa	4	2	6.66(1.15 – 38.71)	**0.035***
Dagaare/wala/sisala[1]	253	19	1	
Educational status				
None	49	2	0.55(0.07 – 4.14)	0.562
Basic	168	13	1.04(0.22 – 4.89)	0.956
Secondary	35	4	1.54(0.26 – 9.06)	0.631
Tertiary[1]	27	2	1	
Occupation				
Student/unemployed	49	4	0.47(0.11 – 2.05)	0.314
Farmer/housewife	83	8	0.55(0.15 – 2.05)	0.368
Business/civil servants	23	1	0.25(0.03 – 2.41)	0.231
Trader	101	4	0.23(0.05 – 0.98)	0.057
Other	23	4	1	

*-P< 0.05- *statistically significant* [1]- *reference group* cOR-*Crude odds ratio*

4.5.2 Obstetric and health service factors associated with placental malaria

At the crude level mothers who took less than three doses of SP had greater odds of placental malaria compared with their counterparts who took three or more doses (cOR=3.46, 95%CI= 1.23 – 9.71). Primigravid mothers were four times more likely of having placental malaria compared with multigravida women (cOR=4.5, 95%CI= 1.79 – 11.29). This study also revealed significant association between ANC visits (cOR= 18.08, 95%CI= 6.79 – 48.13), hypertension (cOR= 12.1, 95%CI= 2.51 – 58.61), malaria in index pregnancy (cOR= 4.1, 95%CI= 1.56 – 11.01) and those who did not have IRS exposure at their homes (cOR= 6.1, 95%CI= 2.44 – 15.27).

Mothers use of ITN and HIV status were not associated with placental malaria from this study. Table 7 shows in detail the obstetric and health service factors associated with placental malaria from this study.

Table 7: Analysis of obstetric and health service factors associated with placental malaria

Characteristics	Placenta malaria		Crude OR (95%CI)	p-value
	No	**Yes**		
IPT-SP				
< 3 doses	134	16	3.46(1.23 – 9.71)	**0.018***
≥ *3 doses*[1]	145	5	1	
Gravidity				
Primigravida	74	13	4.5(1.79 – 11.29)	**<0.0001***
Multigravida[1]	205	8	1	
ITN use				
No	110	12	2.04(0.84 – 5.02)	0.117
Usage[1]	169	9	1	
IRS				
Not expose	50	12	6.1(2.44 – 15.27)	**<0.0001***
Expose[1]	229	9	1	
HIV status				
Negative	273	20	0.43(0.05 – 3.83)	0.457
Positive[1]	6	1	1	
ANC visits				
< 4 visits	23	13	18.08(6.79 – 48.13)	**<0.0001***
≥*4 visits*[1]	256	8	1	
Malaria in index pregnancy				
No	174	6	1	
Yes	105	15	4.1(1.56 – 11.01)	**0.004***
Hypertension				
Normotension	275	17	1	
Hypertension	4	3	12.1(2.51 – 58.61)	**0.002***

*-P< 0.05- *statistically significant* [1]- *reference group* cOR-*Crude odds ratio*

4.6 Multivariate analysis of factors associated with placental malaria

The factors which were significant at the univariate level for both socio-demographic and obstetric as well as health service factors were all used for the multivariate analysis. After adjustment for other variables, gravidity and ANC visits were the only factors significantly associated with placental malaria, (aOR=3.48, 95%CI= 1.01 – 12.01) and (aOR=9.78, 95%CI= 2.89 – 33.11) respectively.

Although mother's ethnicity, marital status and hypertension all had odds greater than two, they were not significantly associated with placental malaria. Other factors such as IPT-SP and IRS were not significant after adjustment. Table 8 shows the p-values and adjusted odds ratio of the factors in the multivariate analysis.

Table 8: Multivariate analysis of factors associated with placental malaria

Characteristics	aOR	95%CI	p-value
Gravidity			
Primigravida	3.48	1.01 – 12.01	**0.049***
Multigravida[1]			
ANC visits			
< 4 attendance	9.78	2.89 – 33.11	**<0.0001***
≥ *4 attendance*[1]			
IPT-SP			
< 3 doses	1.22	0.35 – 4.29	0.758
≥ *3 doses*[1]			
Ethnicity			
Frafra/Kasena/builsa	2.85	0.18 – 45.71	0.460
Dagaare/wala/sisala[1]			
Marital status			
Not Married	2.28	0.42 – 12.22	0.337
Married[1]			
Maternal age			
≤ 20 years	1.01	0.22 – 4.59	0.988
>20 years[1]			
Malaria in index pregnancy			
Yes	1.64	0.47 – 5.69	0.436
No			
Hypertension status			
Hypertension	2.85	0.30 – 26.79	0.360
Normotension			
IRS			
No	1.86	0.49 – 7.11	0.362
Yes			

*-P< 0.05- *statistically significant* aOR-*adjusted odds ratio* [1]- *reference group*

CHAPTER FIVE

DISCUSSION

The study assessed the proportion of women with placental malaria in Upper West Regional Hospital as well as the sociodemographic, obstetric and health service factors associated with placental malaria. The study revealed antenatal care attendance and gravidity to be significantly associated with placental malaria.

5.1 Proportion of women with placental malaria

The study revealed a low proportion of mothers with placental malaria delivering at the Upper West Regional Hospital. This proportion of 7% was similar to findings from Ndeserua et al., (2015) and Kabanywanyi et al., (2008) who both found 8% prevalence of placental malaria among women delivering at two different hospitals in Tanzania.

A cross-sectional hospital based study in Morogoro, Tanzania by Mosha, Ntarukimana, & John, (2010) also revealed 7% of women delivering at the facility with placental malaria. In this study, majority of the women (67%) with placental malaria had parasite density in the range of 501 to 5000 parasites/µl. This is similar to a study conducted by Babalola et al., (2015) where microscopy technique was used to detect placental malaria with about 60% of the positive samples having parasite density in the range of 501 to 5000 parasite/µl. This moderately high parasite density could lead to adverse neonatal outcomes and hence congenital malaria in the newborn.

In contrast to the low percentage of women with placental malaria from this study, a high percentage (52%) was recorded in a prospective study conducted by van Spronsen et al., (2012) in the northern part of Ghana. The two study areas are similar in geographic location, ethnic

background and climate. However, the difference in the prevalence could be due to malaria in pregnancy prevention interventions. Whereas in this study mothers benefitted from five doses of IPT-SP, it was just three doses of IPT-SP that was available for the mothers in the higher prevalent study. It is however not surprising considering the decline comparing both studies as Hommerich et al., (2007) noted a 57% decline in placental malaria in the southern part of Ghana between the years 2000 and 2006.

The low proportion of women with placental malaria could be due to a larger proportion of women receiving two or more doses of IPT-SP. In this study over 80% of the women received two or more doses of SP. Although SP resistance is of increasing concern, it is still effective in preventing malaria during pregnancy as reported by several studies (Tutu, Lawson, & Browne, 2011; WHO, 2007; Wilson et al., 2011).

Another reason for the low proportion of placental malaria could be due to other malaria interventions such as ownership and use of insecticide treated bed nets by mothers and also indoor residual spraying for vector control that has been ongoing in the region. From the results of the study majority of the parturient women used insecticide treated bed nets during the pregnancy period and close to 80% of the study respondents had their homes covered with IRS exercise.

Furthermore, the proportion of women with placental malaria could have been more using diagnostic techniques such as histopathology of the placental tissue instead of blood film microscopy used in this study. Histopathology diagnosis of placental malaria is superior to microscopy in terms of specificity (Kabongo et al., 2016). This study employed blood film microscopy of placental blood for a quicker and simpler way of detecting placental parasitemia. The blood film microscopy technique have been used to detect placental malaria by several

researchers including (Asante et al., 2013; Bako et al., 2009; Mwandama et al., 2015; Ndeserua et al., 2015).

The low proportion of placental malaria means that the associated effect of placental malaria such as LBW, miscarriages, preterm delivery, prematurity, IUGR and maternal anemia in the study area should be reducing. Since placental malaria is associated with poor pregnancy and delivery outcomes, recording low prevalence will contribute to improved maternal and neonatal health.

5.2 Socio-demographic characteristics

A higher proportion of the women were within the age category 25 to 34 years, with over 80% of the women above 20 years. Although mothers who were 20 years or less had a significant increase in odds of placental malaria at the univariate level, this was not significant after adjustment. Other studies in Tanzania and Papua New Guinea by Ndeserua et al., (2015) and (Lufele et al., 2017) respectively found no significant association between maternal age and placental malaria. Similar results were also reported by Ezebialu et al., (2012) where their study found no significant increase in odds among women greater than or less than 30 years.

Conversely, a cross-sectional study conducted in Nigeria reported a four-fold significant increase in odds among women aged 18 to 22 years (Babalola et al., 2015). Thus, maternal age could be a significant factor contributing to placental malaria in some jurisdictions but this was not evident in this study. The study results on age suggest that maternal age should not be the focus when prioritizing interventions for malaria control and prevention among pregnant women in the Upper West Regional Hospital.

In this research, few of the women were not married and this was associated with a significant increase in odds of 12 times of them with placental malaria compared with the married ones at the univariate level. However, adjustment of marital status in the multivariate analysis did not yield significance although those not married were still associated with 2 times increase in odds of having placental malaria. This finding conforms with results from (Bako et al., 2009; Cisse et al., 2016; Elhassan et al., 2017) who all noted no association between marital status and placental malaria. Irrespective of the pregnant woman's marital status, exposure to *Plasmodium* will lead to placental malaria and so there is no protection from either being single or married.

Other socio-demographic factors such as religion, occupation and educational status were not associated with placental malaria at the univariate level. The findings of these three socio-demographic factors are similarly reported in a study by Lufele et al., (2017) who found no association between the educational status of the mother and placental malaria as well as the religious and educational status. The results were not also different from what was found in a study conducted in Blue Nile State of Sudan where there was no association between educational status and placental malaria (Elhassan et al., 2017). In that study, pregnant women who had secondary education or lower had a 50% risk of placental malaria although this was not statistically significant. Similarly, Babalola et al., (2015) reported no significant association between educational status and placental malaria as well as religion and occupational status among Nigerian women.

Since many Africans attach affection to religious affiliations which is often a barrier to overcome during implementation of health intervention, the results of this study showing no association will ensure that no particular religion is targeted as priority for malaria prevention in pregnancy. Hence any intervention will be suggested to target all religious groups equally within the study area.

5.3 Obstetric characteristics

The obstetric characteristics that were found to be significant were gravidity and antenatal care attendance. This study results showed that primigravida has a threefold increase in odds of placental malaria compared with multigravid women. This finding is consistent with several studies detailing the significant association between women who become pregnant for the first time and placental malaria compared with their counterparts entering their second or more pregnancy. A prospective cohort study on women delivering at a hospital in Papua New Guinea documented a two times increased in odds of placenta malaria in primigravid mothers compared with multigravida counterparts (Lufele et al., 2017). Ndeserua, Juma, Mosha, & Chilongola, (2015) also showed a significant higher risk of placental malaria among women in Tanzania in their cross-sectional study. In Nigeria, a study conducted by Babalola et al., (2015) also agreed with this study as results of their study showed primigravid women having a significant increase in odds by two times compared with multigravid mothers.

It is a general consensus from several research by scholars that primigravid or nulliparous women are more susceptible to placental malaria. This stems from the fact that their immune system is being primed for the first time in the pregnancy in relation to *Plasmodium* parasite exposure. Placental malaria occurs when the *Plasmodium* infested red blood cell bind to chondroitin sulfate A (CSA) in the intervillous regions of the placenta leading to sequestration (Gaw et al., 2018). In secundigravida and multigravida, previous exposure to *Plasmodium* infection resulted in the production of antibodies that will bind to Chondroitin sulfate A (CSA) in subsequent pregnancies thereby interfering with adhesion of infected *Plasmodium* red blood cells to CSA (Magistrado et al., 2008). This phenomenon reduces the risk of placental malaria in multigravid women.

Knowledge on primigravida being strongly associated with placenta malaria will help health educators emphasize the need for mothers being pregnant for the first time to regularly and timely attend antenatal services as well as adhere to malaria prevention and control strategies.

Antenatal care attendance is very important as it provides an avenue for mothers who are pregnant for the first time to be educated on what to expect during the pregnancy and also those with their second or more pregnancy to share their experience with the new ones and also be educated as well. For this study, majority (88%) of the pregnant women attended antenatal care four or more times as recommended by WHO for normal pregnancy. This attendance percentage is not far different from the 78% reported by Fondjo et al., (2018) in a cross-sectional study among pregnant women in Kumasi, Ghana. Similarly, another cross-sectional study in Ghana reported 90% of women attending at least four ANC visits (Asundep et al., 2013). The relatively high ANC attendance in the regional hospital could be because of accessibility as well as it being urbanely situated. The figure could have been lower in rural communities.

The high ANC attendance by pregnant women offers an opportunity for government and other malaria stakeholders to implement malaria prevention strategies at the ANC level to successfully control malaria in pregnancy. It also offers the opportunity for health care providers to effectively manage pregnancy related complications in order to reduce maternal mortality.

From this study, statistical significance association was found among women attending less than four antenatal visits and placental malaria. These women had an increased risk of nine times the odds of having placental malaria compared with women who met the WHO recommended visits. A study conducted in Sudan also reported a higher odds among women who did not meet the recommended WHO ANC visit to having placental malaria (Elhassan et al., 2017). This similar finding suggest that ANC attendance is very important in efforts aimed at controlling malaria in

pregnancy and so advocacy should be targeted at all pregnant women attending at least four ANC visits before delivery. In order to achieve this 100% target, pregnant women should book early to attend ANC during the pregnancy. Booking early and attending more visit will enable them receive all the recommended doses of IPT-SP. Since IPT-SP is a directly observed therapeutic drug, the pregnant woman must be at the health facility to receive the drug.

Other factors such as ITN use, IPT-SP, IRS, HIV status and malaria in index pregnancy were not significant at the multivariate level. The results of IPT-SP insignificantly associated with placental malaria is astonishing as many research have demonstrated the benefits of IPT-SP in preventing malaria in pregnancy (Hommerich et al., 2007; Masaninga et al., 2016). The finding from this study is however not different from what Ndeserua et al., (2015), van Spronsen et al., (2012) and Harrington, Mutabingwa, Kabyemela, Fried, & Duffy, (2011) found in their studies. The possible reasons why IPT-SP was not associated with placental malaria could be that, the health care provider possibly did not directly observe the women taking the drug and so like what was reported in some studies where the women actually spit it out after the health care provider moves away or not paying attention. Sulfadoxine pyrimethamine resistance is becoming increasingly widespread especially in malaria endemic countries (Mbonye et al., 2015) and this could possibly explain the insignificant association in this study.

For ITN use, findings from a study in Nigeria showed a two fold increase in association between those who do not use ITN and placental malaria compared with women who use it (Babalola et al., 2015). Another study by Ezebialu et al., (2012) found significant association between ITN use and placental malaria. These results do not corroborate with what was found in this study where no association was established.

Even though the mothers reported to having slept under the mosquito net, this was not verified by observation and so it is possible that some of those who reported sleeping under the net did not actually do so. Also, those who slept under the net may have used torn or pierced nets not treated and hence trapping mosquitoes in it.

The mothers HIV status did not pose any risk to having placental malaria as reported in this study.The finding is consistent to what was found by Ezebialu et al., (2012). In contrast to this finding, a study on Sub-Saharan African women reported statistically significant association between HIV positive mothers and placental malaria (Uneke, 2007). These women had an increased risk of placental malaria compared with HIV negative women. The reason for HIV status of the mother not associated with placental malaria in this study could be due to the lower proportion of mothers (4.7%) and the fact that IPT-SP is contraindicated in HIV mothers receiving cotrimoxazole.

5.4 Environmental characteristics

Indoor residual spraying is gaining prominence as it is increasingly evident in reducing malaria in endemic countries. Some studies have presented results of reduced malaria infection in areas that employed IRS as a vector control intervention (Ogouyèmi-hounto et al., 2018). This study did not find any significant association between pregnant women who responded to having their homes sprayed in the course of their pregnancy and placental malaria. This finding is, however, different from what Muhindo et al., (2016) found where IRS was significantly associated with a reduction in placental malaria prevalence in women who had IRS protection. The possible explanation for the no association between IRS and placental malaria could be that, some of the women who responded to having IRS exposure may not have had IRS exposure since we could not verify this.

Despite this finding, combining IRS with other malaria control intervention would help decrease the prevalence of placental malaria as it has been demonstrated in other studies.

Living in a congested apartment such as more than two occupants per living room increase the risk of placental malaria by 60% (Babalola et al., 2015). This study did not however find significant association between number of occupants in a living room and placental malaria. Although overcrowding and frequent opening of living room door has been the reason for increased risk of malaria (Lindsay et al., 2003), it is possible that this study participants who lived with more than two people in the same room did not frequently open their doors and hence were not exposed to mosquito bites often.

5.5 Limitations of study

This was a cross-sectional study done in the regional hospital; hence, the findings are not generalizable. The data was not collected in the peak malaria season and so could have affected the proportion of women with placental malaria. Social desirability could have affected some of the responses provided by the participants.

CHAPTER SIX

CONCLUSIONS AND RECOMMENDATIONS

6.1 Conclusion

The proportion of women with placental malaria who delivered at the Upper West Regional Hospital was low (7%) with *Plasmodium falciparum* accounting for the placental malaria infection.

Primigravida and less than four antenatal care visits were significantly associated with placental malaria after controlling for other obstetric factors such as IPT-SP intake, ITN use, hypertension as well as socio-demographic factors such as maternal age and ethnicity. Both factors were associated with increased odds of placental malaria.

Although women receiving less than 3 doses of IPT-SP was associated with a 22% increase risk of placental malaria after controlling for obstetric and socio-demographic factors, this was not statistically significant.

There was no significant association between ITN use and placental malaria.

All the sociodemographic factors were not associated with placental malaria after adjustment, however maternal age 20 years or less and mothers who were not married had increased risk of placental malaria at the crude level.

6.2 Recommendations

The Public Health Unit of the hospital and the municipal health directorate should organize community education and sensitization programs within the municipality on the importance of ANC attendance so as to increase the number of ANC visits by pregnant mothers. This will ensure that all women attend more than four ANC visits before delivery so as to reduce their risk of PM.

Antenatal care providers should encourage male partner involvement especially with primigravid women as this will help increase the number of ANC visits by these women before delivery.

Primigravid women should be given special attention during their ANC visits by health care providers and encouraged to sleep under treated bed nets.

REFERENCES

Anchang-Kimbi, J. K., Achidi, E. A., Apinjoh, T. O., Mugri, R. N., Chi, H. F., Tata, R. B., … Troye-Blomberg, M. (2014). Antenatal care visit attendance, intermittent preventive treatment during pregnancy (IPTp) and malaria parasitaemia at delivery. *Malaria Journal*, *13*(1), 1–9. https://doi.org/10.1186/1475-2875-13-162

Asante, K. P., Owusu-Agyei, S., Cairns, M., Dodoo, D., Boamah, E. A., Gyasi, R., … Chandramohan, D. (2013). Placental Malaria and the Risk of Malaria in Infants in a High Malaria Transmission Area in Ghana: A Prospective Cohort Study. *The Journal of Infectious Diseases*, *208*(9), 1504–1513. https://doi.org/10.1093/infdis/jit366

Asundep, N. N., Carson, A. P., Turpin, C. A., Tameru, B., Agidi, A. T., Zhang, K., & Jolly, P. E. (2013). Determinants of access to antenatal care and birth outcomes in Kumasi, Ghana. *Journal of Epidemiology and Global Health*, *3*(4), 279–288. https://doi.org/10.1016/j.jegh.2013.09.004

Ayres Pereira, M., Mandel Clausen, T., Pehrson, C., Mao, Y., Resende, M., Daugaard, M., … Salanti, A. (2016). Placental Sequestration of Plasmodium falciparum Malaria Parasites Is Mediated by the Interaction Between VAR2CSA and Chondroitin Sulfate A on Syndecan-1. *PLoS Pathogens*, *12*(8), 1–26. https://doi.org/10.1371/journal.ppat.1005831

Babalola, A. S., Idowu, O. A., Wobo, S. O. S., & Fabusoro, E. (2015). Risk factors associated with occurrence of placental malaria in a population of parturients in Abeokuta , Ogun State , Nigeria. *Malaria Journal*, *6*(8), 1–6.

Bako, B. G., Audu, B. M., Geidam, A. D., Kullima, A. A., Ashiru, G. M., Malah, M. B., … Musa, A. B. (2009). Prevalence, risk factors and effects of placental malaria in the UMTH,

Maiduguri, North-eastern, Nigeria: a cross-sectional study. *Journal of Obstetrics and Gynaecology : The Journal of the Institute of Obstetrics and Gynaecology*, *29*(4), 307–310. Retrieved from http://www.ncbi.nlm.nih.gov/pubmed/19835497

Bassey, G., Nyengidiki, T. K., & John, C. T. (2015). Prevalence of placenta Plasmodium parasitemia and pregnancy outcome in asymptomatic patients at delivery in a University Teaching Hospital in Nigeria. *Nigerian Journal of Clinical Practice*, *18*(1), 27–32. https://doi.org/10.4103/1119-3077.146975

Boateng, I. O., & Anto, F. (2017). Intermittent preventive treatment of malaria in pregnancy : a cross - sectional survey to assess uptake of the new sulfadoxine – pyrimethamine five dose policy in Ghana. *Malaria Journal*, 1–9. https://doi.org/10.1186/s12936-017-1969-7

Bouyou-akotet, M. K., Ionete-collard, D. E., Mabika-, M., Kendjo, E., Matsiegui, P., Mavoungou, E., & Kombila, M. (2003). Prevalence of Plasmodium falciparum infection in pregnant women in Gabon. *Malaria Journal*, *7*, 1–7.

Campos, I. M., Uribe, M. L., Cuesta, C., Franco-Gallego, A., Carmona-Fonseca, J., & Maestre, A. (2011). Diagnosis of gestational, congenital, and placental malaria in colombia: Comparison of the efficacy of microscopy, nested polymerase chain reaction, and histopathology. *American Journal of Tropical Medicine and Hygiene*, *84*(6), 929–935. https://doi.org/10.4269/ajtmh.2011.10-0507

Cisse, M., Diallo, A. H., Some, D. A., PODA, A., Awandare, A. G., & Guiguemde, T. R. (2016). Association of placental Plasmodium falciparum parasitaemia with maternal and newborn outcomes in the periurban area of Bobo-Dioulasso, Burkina Faso. *Parasitology Open*, *2*(2016), e15. https://doi.org/10.1017/pao.2016.12

Cox, S. E., Staalsoe, T., Arthur, P., Bulmer, J. N., Tagbor, H., Hviid, L., … Kirkwood, B. R. (2005). Maternal vitamin A supplementation and immunity to malaria in pregnancy in Ghanaian primigravids. *Tropical Medicine & International Health*, *10*(12), 1286–1297. https://doi.org/10.1111/j.1365-3156.2005.01515.x

Elhassan, M. O., Adam, I., Idress, H. E., Omer, S. A., Noureldein, A. N., Sulaiman, S. M., … Abdelrahim, M. (2017). Placental malaria and its effect on pregnancy outcomes in Sudanese women from Blue Nile State. *Malaria Journal*, *16*(1), 1–8. https://doi.org/10.1186/s12936-017-2028-0

Ezebialu, I. U., Eke, A. C., Ezeagwuna, D. A., Nwachukwu, C. E., Ifediata, F., & Ezebialu, C. U. (2012). Prevalence, pattern, and determinants of placental malaria in a population of southeastern Nigerian parturients. *International Journal of Infectious Diseases*, *16*(12), e860–e865. https://doi.org/10.1016/j.ijid.2012.07.014

Fehintola, A., Fehintola, F., Loto, O., Fasubaa, O., Bakare, B., & Ogundele, O. (2016). Pregnancy and fetal outcome of placental malaria parasitemia in Ile-Ife, Nigeria. *Tropical Journal of Obstetrics and Gynaecology*, *33*(3), 310. https://doi.org/10.4103/0189-5117.199811

Fondjo, L. A., Sarbeng, K., Arthur, S. N., Addai-Mensah, O., Annani-Akollor, M. E., Anto, E. O., & Owiredu, E.-W. (2018). Regular Antenatal Attendance and Education Influence the Uptake of Intermittent Preventive Treatment of Malaria in Pregnancy: A Cross-Sectional Study at the University Hospital, Kumasi, Ghana. *Journal of Tropical Medicine*, *2018*, 1–8. https://doi.org/10.1155/2018/5019215

Gaw, S. L., Ozarslan, N., Montoya, D. J., Robinson, J., Dorsey, G., Feeney, M., … Fisher, S. J.

(2018). 862: Gene expression profiling of fetal Hofbauer cells and maternal intervillous macrophages in placental malaria. *American Journal of Obstetrics and Gynecology*, *218*(1), S513–S514. https://doi.org/10.1016/J.AJOG.2017.11.399

Graffeo, R., Masucci, L., Bugli, F., Damiano, F., Pinnetti, C., Federico, G., & Fadda, G. (2008). Multiple malaria infection in a pregnant woman from Nigeria: Detection by multiplex PCR. *New Microbiologica*, *31*(4), 565–567. https://doi.org/10.1109/TIE.2014.2321342

Griffin, J. B., Lokomba, V., Landis, S. H., Thorp, J. M., Herring, A. H., Tshefu, A. K., … Meshnick, S. R. (2012). Plasmodium falciparum parasitaemia in the first half of pregnancy, uterine and umbilical artery blood flow, and foetal growth: A longitudinal Doppler ultrasound study. *Malaria Journal*, *11*, 1–10. https://doi.org/10.1186/1475-2875-11-319

Harrington, W. E., Mutabingwa, T. K., Kabyemela, E., Fried, M., & Duffy, P. E. (2011). Intermittent Treatment to Prevent Pregnancy Malaria Does Not Confer Benefit in an Area of Widespread Drug Resistance. *Clinical Infectious Diseases*, *53*(3), 224–230. https://doi.org/10.1093/cid/cir376

Harrington, Whitney E., Mutabingwa, T. K., Kabyemela, E., Fried, M., & Duffy, P. E. (2011). Intermittent treatment to prevent pregnancy malaria does not confer benefit in an area of widespread drug resistance. *Clinical Infectious Diseases*, *53*(3), 224–230. https://doi.org/10.1093/cid/cir376

Hommerich, L., Von Oertzen, C., Bedu-Addo, G., Holmberg, V., Acquah, P. A., Eggelte, T. A., … Mockenhaupt, F. P. (2007). Decline of placental malaria in southern Ghana after the implementation of intermittent preventive treatment in pregnancy. *Malaria Journal*, *6*, 1–8. https://doi.org/10.1186/1475-2875-6-144

Ibanga, G. J., Abasiattai, A. M., Bassey, E. A., Ukpe, M., Olatunbosun, O. A., & Ekrikpo, U. (2015). Placental malarial parasitaemia and pregnancy outcome among parturients in a tertiary hospital in South- South Nigeria. *Asian Journal of Medical Sciences*, *6*(6), 53–59. https://doi.org/10.3126/ajms.v6i6.12401

Kabanywanyi, A. M., MacArthur, J. R., Stolk, W. A., Habbema, J. D. F., Mshinda, H., Bloland, P. B., … Kachur, S. P. (2008). Malaria in pregnant women in an area with sustained high coverage of insecticide-treated bed nets. *Malaria Journal*, *7*, 1–7. https://doi.org/10.1186/1475-2875-7-133

Kabongo, J.-M. M., Muehlenbachs, A., Liu, Y., Landis, S. H., Sullivan, D. J., Meshnick, S. R., … Griffin, J. B. (2016). Diagnosis of placental malaria in poorly fixed and processed placental tissue. *Malaria Journal*, *15*(1), 1–6. https://doi.org/10.1186/s12936-016-1314-6

Karlsen, S., Say, L., Souza, J., Hogue, C. J., Calles, D. L., Gülmezoglu, A. M., & Raine, R. (2011). The relationship between maternal education and mortality among women giving birth in health care institutions : Analysis of the cross sectional WHO Global Survey on Maternal and Perinatal Health, 1–10.

Lindsay, S. W., Jawara, M., Paine, K., Pinder, M., Walraven, G. E. L., & Emerson, P. M. (2003). Changes in house design reduce exposure to malaria mosquitoes. *Tropical Medicine and International Health*, *8*(6), 512–517. https://doi.org/10.1046/j.1365-3156.2003.01059.x

Lufele, E., Umbers, A., Ordi, J., Ome-Kaius, M., Wangnapi, R., Unger, H., … Rogerson, S. (2017). Risk factors and pregnancy outcomes associated with placental malaria in a prospective cohort of Papua New Guinean women. *Malaria Journal*, *16*(1), 427. https://doi.org/10.1186/s12936-017-2077-4

Magistrado, P., Salanti, A., Tuikue Ndam, N. G., Mwakalinga, S. B., Resende, M., Dahlbäck, M., … Nielsen, M. A. (2008). VAR2CSA Expression on the Surface of Placenta-Derived *Plasmodium falciparum* –Infected Erythrocytes. *The Journal of Infectious Diseases*, *198*(7), 1071–1074. https://doi.org/10.1086/591502

Masaninga, F., Katepa Bwalya, M., Malumo, S., Hamainza, B., Songolo, P., Kamuliwo, M., … Babaniyi, O. A. (2016). Increased uptake of intermittent preventive treatment for malaria in pregnant women in Zambia (2006–2012): Potential determinants and highlight of lessons learnt. *Asian Pacific Journal of Tropical Biomedicine*, *6*(7). https://doi.org/10.1016/j.apjtb.2016.01.010

Mbonye, A. K., Birungi, J., Yanow, S. K., Shokoples, S., Malamba, S., Alifrangis, M., & Magnussen, P. (2015). Prevalence of Plasmodium falciparum Resistance Markers to Sulfadoxine-Pyrimethamine among Pregnant Women Receiving Intermittent Preventive Treatment for Malaria in Uganda. *Antimicrobial Agents and Chemotherapy*, *59*(9), 5475–5482. https://doi.org/10.1128/aac.00507-15

Menendez, C., Ordi, J., Ismail, M. R., Ventura, P. J., Aponte, J. J., Kahigwa, E., … Alonso, P. L. (2000). The Impact of Placental Malaria on Gestational Age and Birth Weight. *The Journal of Infectious Diseases*, *181*(5), 1740–1745. https://doi.org/10.1086/315449

Mockenhaupt, F. P., Bedu-Addo, G., Von Gaertner, C., Boyé, R., Fricke, K., Hannibal, I., … Bienzle, U. (2006). Detection and clinical manifestation of placental malaria in southern Ghana. *Malaria Journal*, *5*, 1–10. https://doi.org/10.1186/1475-2875-5-119

Mohammed, B. H., Johnston, J. M., Vackova, D., Hassen, S. M., & Yi, H. (2019). The role of male partner in utilization of maternal health care services in Ethiopia : a community-based

couple study. *BMC Pregnancy and Childbirth*, 1–9.

Moore, K. A., Simpson, J. A., Paw, M. K., Pimanpanarak, M. P. J., Wiladphaingern, J., Rijken, M. J., … McGready, R. (2016). Safety of artemisinins in first trimester of prospectively followed pregnancies: An observational study. *The Lancet Infectious Diseases*, *16*(5), 576–583. https://doi.org/10.1016/S1473-3099(15)00547-2

Mosha, T. C. E., Ntarukimana, D., & John, M. (2010). Prevalence of congenital malaria among neonates at Morogoro Regional Hospital, Morogoro, Tanzania. *Tanzania Journal of Health Research*, *12*(4), 241–248. Retrieved from http://www.ncbi.nlm.nih.gov/pubmed/24409631

Muhindo, M. K., Kakuru, A., Natureeba, P., Awori, P., Olwoch, P., Ategeka, J., … Jagannathan, P. (2016). Reductions in malaria in pregnancy and adverse birth outcomes following indoor residual spraying of insecticide in Uganda. *Malaria Journal*, *15*(1), 1–9. https://doi.org/10.1186/s12936-016-1489-x

Mwandama, D., Gutman, J., Wolkon, A., Luka, M., Jafali, J., Ali, D., … Skarbinski, J. (2015). The use of intermittent preventive treatment in pregnancy and insecticide-treated bed nets for malaria prevention by women of child-bearing age in eight districts in Malawi. *Malaria Journal*, *14*(1), 1–10. https://doi.org/10.1186/s12936-015-0840-y

Ndeserua, R., Juma, A., Mosha, D., & Chilongola, J. (2015a). Risk factors for placental malaria and associated adverse pregnancy outcomes in Rufiji, Tanzania: A hospital based cross sectional study. *African Health Sciences*, *15*(3), 810–818. https://doi.org/10.4314/ahs.v15i3.15

Ndeserua, R., Juma, A., Mosha, D., & Chilongola, J. (2015b). Risk factors for placental malaria and associated adverse pregnancy outcomes in Rufiji , Tanzania : a hospital based cross

sectional study ., *15*(3), 810–818.

Nsibu, C. N., Manianga, C., Kapanga, S., Mona, E., Pululu, P., & Aloni, M. N. (2016). Determinants of Antenatal Care Attendance among Pregnant Women Living in Endemic Malaria Settings: Experience from the Democratic Republic of Congo. *Obstetrics and Gynecology International*, *2016*. https://doi.org/10.1155/2016/5423413

Nwaefuna, E. K., Afoakwah, R., Orish, V. N., Egyir-yawson, A., & Boampong, J. N. (2015). Effectiveness of Intermittent Preventive Treatment in Pregnancy with Sulphadoxine-Pyrimethamine against Submicroscopic falciparum Malaria in Central Region , Ghana. *Malaria Journal*, *2015*. https://doi.org/10.1155/2015/959427

Ofori, M. F., Ansah, E., Agyepong, I., Ofori-Adjei, D., Hviid, L., & Akanmori, B. D. (2009). Pregnancy-associated malaria in a rural community of Ghana. *Ghana Medical Journal*, *43*(1), 13–18.

Ofori, M. F., Lamptey, H., Dickson, E. K., Kyei-Baafour, E., & Hviid, L. (2018). Etiology of Placental Plasmodium falciparum Malaria in African Women. *The Journal of Infectious Diseases*, *218*(2), 277–281. https://doi.org/10.1093/infdis/jiy168

Ogouyèmi-hounto, A., Hounkpe, B., Sissinto, Y., Tove, S. De, Houngbegnon, P., Koukpoliyi, A., … Gazard, D. K. (2018). Effect of indoor residual spraying on malaria incidence , parasitemia and parasite density among children less than five years of age in northern Benin. *Journal of Paraitology and Vector Biology*, *10*(October), 129–137. https://doi.org/10.5897/JPVB2018.0342

Okell, L. C., Griffin, J. T., & Roper, C. (2017). Mapping sulphadoxine- Plasmodium falciparum malaria in infected humans and in parasite populations in Africa. *Scientific Reports*,

(November 2016), 1–15. https://doi.org/10.1038/s41598-017-06708-9

Oraneli, B. U., Okeke, O. C., & Ubachukwu, P. O. (2013). Effect of placental malaria on birth weight of babies in Nnewi, Anambra state, Nigeria. *Journal of Vector Borne Diseases*, *50*(1), 13–17.

Pekyi, D., Ampromfi, A. A., Tinto, H., Traoré-Coulibaly, M., Tahita, M. C., Valéa, I., … Mulenga, J. (2016). Four artemisinin-based treatments in African pregnant women with malaria. *Malawi Medical Journal*, *28*(3). https://doi.org/10.1056/NEJMoa1508606

Rachas, A., Le Port, A., Cottrell, G., Guerra, J., Choudat, I., Bouscaillou, J., … Garcia, A. (2012). Placental malaria is associated with increased risk of nonmalaria infection during the first 18 months of life in a beninese population. *Clinical Infectious Diseases*, *55*(5), 672–678. https://doi.org/10.1093/cid/cis490

Ruragiriwa, A. A., Mogren, I., Nyirazinyoye, L., Ntaganira, J., & Krantz, G. (2017). Determinants of Poor Utilization of Antenatal Care Services among Recently Delivered Women in Rwanda: A Population Based Study. *MCN The American Journal of Maternal/Child Nursing*, *42*(6), 366. https://doi.org/10.1097/NMC.0000000000000382

Schantz-Dunn, J., & Nour, N. M. (2009). Malaria and Pregnancy A Global Health Perspective. *Rev Obstet Gynecol.*, *2*(3), 1–2. https://doi.org/10.3909/riog0091

Sharma, L., & Shukla, G. (2017). Placental Malaria: A New Insight into the Pathophysiology. *Frontiers in Medicine*, *4*, 117. https://doi.org/10.3389/fmed.2017.00117

Singh, J., Soni, D., Mishra, D., Singh, H. P., & Bijesh, S. (2014). Placental and neonatal outcome in maternal malaria. *Indian Pediatrics*, *51*(4), 285–288. https://doi.org/10.1007/s13312-014-

0402-3

Stephens, J. K., Baafour, E. K., Dickson, E. K., Ofori, J. K., Ofori, M. F., Wilson, M. L., … Akanmori, B. D. (2017). Effect of IPTp on Plasmodium falciparum antibody levels among pregnant women and their babies in a sub - urban coastal area in Ghana. *Malaria Journal*, 1–10. https://doi.org/10.1186/s12936-017-1857-1

Tako, E. A., Zhou, A., Lohoue, J., Leke, R., Taylor, D. W., & Leke, R. F. G. (2005). Risk Factors for Placental Malaria and Its Effect on Pregnancy Outcome in Yaounde, Cameroon. *The American Journal of Tropical Medicine and Hygiene*, *72*(3), 236–242. https://doi.org/10.4269/ajtmh.2005.72.236

Tuladhar, H., & Dhakal, N. (2012). Impact of Antenatal Care on Maternal and Perinatal utcome: A Study at Nepal Medical College Teaching Hospital. *Nepal Journal of Obstetrics and Gynaecology*, *6*(2), 37–43. https://doi.org/10.3126/njog.v6i2.6755

Tutu, E. O., Lawson, B., & Browne, E. (2011). The effectiveness and perception of the use of sulphadoxine-pyrimethamine in intermittent preventive treatment of malaria in pregnancy programme in Offinso district of ashanti region , Ghana. *Malaria Journal*, *10*(1), 385. https://doi.org/10.1186/1475-2875-10-385

Uneke, C. J. (2007). Impact of placental Plasmodium falciparum malaria on pregnancy and perinatal outcome in sub-Saharan Africa: I: introduction to placental malaria. *The Yale Journal of Biology and Medicine*, *80*(2), 39–50. https://doi.org/10.1007/s00256-010-0904-6

Uneke, C. J. (2008). Impact of placental Plasmodium falciparum malaria on pregnancy and perinatal outcome in sub-Saharan Africa. Part III: Placental Malaria, Maternal Health, and Public Health. *Yale Journal of Biology and Medicine*, *81*(1), 1–7.

van Spronsen, J. H. M., Schneider, T. A. J., & Atasige, S. (2012). Placental malaria and the relationship to pregnancy outcome at Gushegu District Hospital, Northern Ghana. *Tropical Doctor*, *42*(2), 80–84. https://doi.org/10.1258/td.2011.110431

Wanjiku, G. (2011). EliScholar – A Digital Platform for Scholarly Publishing at Yale Low Uptake Of Intermittent Preventive Treatment Of Malaria In Hiv-Infected Pregnant Women At An Urban Hospital In Accra , Ghana. *Yale Medicine Thesis Digital Library*, (January). Retrieved from http://elischolar.library.yale.edu/ymtdl/1604

WHO. (2007). WHO Guidance for Prevention and Treatment of Malaria in Pregnancy Insecticide-treated nets (ITNs) Sources: 1. A Strategic Framework for Malaria Prevention and Control during Pregnancy in the African Region, WHO 2004. 2. WHO Global Malaria Programme: Posit. Retrieved from https://www.mchip.net/sites/default/files/03. WHO Guidance for MIP Prevention and Treatment.pdf

WHO. (2012). *WHO Evidence Review Group: Intermittent Preventive Treatment of malaria in pregnancy (IPTp) with Sulfadoxine-Pyrimethamine (SP)* (Vol. 1).

WHO. (2016). *WHO recommendations on antenatal care for a positive pregnancy experience*. Retrieved from http://www.who.int

Wilson, N. O., Ceesay, F. K., Obed, S. A., Adjei, A. A., Gyasi, R. K., Rodney, P., … Stiles, J. K. (2011). Intermittent preventive treatment with sulfadoxine-pyrimethamine against malaria and anemia in pregnant women. *American Journal of Tropical Medicine and Hygiene*, *85*(1), 1–21. https://doi.org/10.4269/ajtmh.2011.10-0512

Appendices

Appendix A: Consent form and Respondents information

Title of study	Factors associated with placental malaria in Upper West Regional Hospital
Objective	To assess the prevalence of factors associated with Placental malaria in Upper West Regional Hospital
Name of Principal investigator	Pascal Kingsley Mwin
Name of Supervisor	Dr Frederick Wurapa & Prof. Col.(Rtd) Edwin Afari
Information for participants	I am an MPhil student of School of Public Health, University of Ghana- Legon. I am conducting a study to assess the prevalence of factors associated with placental in partial fulfill of the requirement of a master of Philosophy in Applied Epidemiology and Disease Control. Your participation and honest information in this study will be most appreciated.

Procedure	You will be asked some few questions on your pregnancy, antenatal care and malaria. Your responses to these questions will be ticked appropriately on the questionnaire. Your Antenatal record book will also be reviewed to fill in some of the questions. When you deliver, blood sample from your placenta will be taken for testing to find out if there are malaria parasites in the sample
Risk	There is no direct risk in participating in this study. You may however be required to answer few questions from research assistants.

Benefits	There are no direct benefits from this study. However, results from this study will be used to prioritize malaria intervention implementations in other to prevent malaria in pregnancy
Confidentiality	Be assured that all information obtained from you will be treated with utmost confidentiality and used strictly for the purposes of the study. You will not be associated with the information provided during the study.
Right to refuse	You have the right to refuse to take part in this study. You may also freely withdraw at any point in the study.
Consent	The content of this form and the purpose of the study and the risk and benefits have been read and explained to me in the language I understand. I do hereby give my consent to participate in the study.
Signature/thumb print of participant	
Date	

Signature of Investigator	
Date	
Contact	

For any enquiries, further questions or additional information, please contact the Principal investigator on Email: mwinpascalkingsley@yahoo.com Mobile 0577680050

Or Miss Hannah Frimpong, GHS Ethics Review Committee on 0244094752, Email: Hanna.frimpong@ghsmail.org

Appendix B: Questionnaire

Factors associated with Placental Malaria in Upper West Regional Hospital.

Instruction: Tick the relevant option and write in the space provided on the right

Introduction

I am Pascal Kingsley Mwin of the School of Public Health, University of Ghana with a team of researchers. We are conducting a study on factors that are associated with placental malaria in this Hospital. You will be required to answer a few questions about yourself and malaria during this pregnancy.

The data collected will be kept confidential. The findings will be helpful in putting measures in place to control malaria in pregnancy. The interview will last about 10 – 15 minutes.

……..

Date of Interview

Consent form must be checked before starting

PART A: BASIC INFORMATION

1.1 Maternal Study ID					
1.2 Name of RA…….					
1.3 Has consent been given?				1. Yes	2. No

If consent is not given, cross out the rest of the form after writing reason for refusal.

PART B: SOCIO-DEMOGRAPHIC CHARACTERISTICS

1. Maternal Age (Years) ……
2. Marital Status: Single [ ] Married [ ] Divorced[ ] Co-habiting [ ]
   Widowed [ ]
3. Educational Level: None [ ]Primary [ ] Middle School [ ] JHS [ ]
   SHS [ ] Tertiary [ ]
4. Ethnicity: Dagaare/Wala/Sisala [ ] Frafra/Kasena/Builsa [ ]
   Dagomba/Gonja/Mamprusi [ ] Akan [ ]
   Fante [ ] Ewe [ ] Other [ ] Specify……
5. Religion: Christianity [ ] Islam [ ] African Traditional [ ]
   Others [ ]

6. Occupation: Student/Unemployed [ ] Farmer/House wife[ ]

Business or Civil servant[ ] Trader [ ] Other [ ] Specify……

PART C: SOCIOECONOMIC

7. Do you own any land Yes [ ] No [ ]

8. Do you have land on which you farm?

Yes, my own [ ] Yes, part of family land [ ] Yes part of husband's [ ]

Yes, rented land [ ] Yes, rented land [ ] No [ ]

9. Which crops do you mainly grow on your land?

Food items, mainly for home consumption [ ]

Food items, mainly for sale on the market [ ]

Cash crops [ ]

PART D: THE HOUSE/SURROUNDINGS

Say "now I am going to ask you about your 'household & surrounding'". Explain to subject what a household is.

10. How many of you sleep in the room that you have slept in during this pregnancy?

Two people [ ] Three people [ ] Four people [ ] Five and above [ ]

11. Does your house have electricity? Yes [ ] No [ ]

12. Is the household part of a compound?

No, free standing [ ] Common wall, but not enclosed [ ]

courtyard with more than 1 entrance [ ]

courtyard with only 1 entrance [ ]

13. What is the main source of drinking water for members of your household?

Piped into home/compound [ ] Public tap [ ]

Hand pump/ closed bore hole [ ] Closed well [ ]

Open well [ ] Stream/ river [ ] Lake/dam/pond [ ]

Rain water [ ] Sachet/ "Pure water" [ ] Bottled water [ ]

Other [ ], specify…….....

14. What kind of toilet facility does your household have?

1. Flush latrine / WC [ ] 2. Ventilated improved pit (VIP) /KVIP [ ]

3. Other pit latrine [ ] 4. Open fields [ ]

5. Defaecates in house but faeces transferred elsewhere / bucket latrine [ ]

6. Other Specify [ ]…….....

15**.** Do you own/rent the house you live in, or have another type of arrangement, such as "perching"?

1. Sole Ownership [ ] 2. Joint Ownership [ ] 3. Family/relation's house [ ]

4. House provided rent free [ ] 5. Renting [ ] 6. Perching [ ]

7. Other Specify [ ]……

16**.** Is the surrounding of your house bushy Yes [ ] No [ ]

ITN / INSECTICIDE / ANTIMALARIAL DRUG USE

17. Do you have an insecticide-treated bed net (ITN)? Yes [ ] No [ ]

Skip 39 & 40 if answer No

18. Did you sleep under an ITN in the last 7 nights? Yes [ ] No [ ]

19. Is the ITN pierced/torn? Yes [ ] No [ ]

20. Have you used any of the following in the last 7 days?

Mosquito coil	Yes [ ]	No [ ]
Insecticide sprays	Yes [ ]	No [ ]
Commercial repellents	Yes [ ]	No [ ]
Traditional repellents	Yes [ ]	No [ ]

21. Have your house been sprayed (IRS) by any organization during the period of your pregnancy? Yes [ ] No [ ]

22. Have you been ill with a fever in the last 24 hours? Yes [ ] No [ ]

23. Have you been treated for malaria during this pregnancy? Yes [ ] No [ ]

24. Have you taken any antimalarial drug in the last 14 days? Yes [ ] No [ ]

CHECKLIST FOR MATERNAL INFORMATION FROM MATERNAL & CHILD RECORD BOOK

PART E: OBSTETRIC HISTORY/ANC VISITS

1.	Total Number of ANC Visits									
2.	What is the number of this pregnancy (Gravidity)?									
3.	How many previous deliveries have you had (Parity)?									
4.	Number of multiple births									
5.	Number of abortions									
6.	Number of caesarean sections									
7.	Last Menstrual Period (LMP), enter 88 888 8888 if Not Known									
8.	Gestational Age (in Weeks)									
9.	Expected Delivery Date (EDD)									
10.	Gestational age determined by	Ultrasound [ ]		2 Fundal height [ ]						

MEASUREMENTS / SCREENING TESTS (AT FIRST ANC VISIT)

11.	Weight (in kg)			.
12.	Height (in meters)			.
13.	Temperature (in ^{o}C)			.
14.	Systolic Blood Pressure (mm Hg)			
15.	Diastolic Blood Pressure (mm Hg)			
16.	VDRL Test	Reactive [ ]	Negative [ ]	Not Done [ ]
17.	Retro Screening	Positive [ ]	Negative [ ]	Not Done [ ]

18.	Sickling Status	Positive [ ]		Negative [ ]		Not Done [ ]
19.	Haemoglobin genotype	AA [ ]		AS [ ]	AC [ ]	SC [ ]
		CC [ ]		SS [ ]		
20.	G6PD Screen	Normal [ ]	Partial [ ]	Deficient [ ]		Not Done [ ]

21. Was SP DOT given taken during this pregnancy? Yes [ ] No [ ]

22. Number of doses of SP given.

One (1) [ ] Two (2) [ ] Three (3) [ ]

Four (4) [ ] Five (5) [ ]

PART F: PREGNANCY OUTCOME /DELIVERY

23.	Date of delivery:								
24.	Gestational age at delivery (in weeks)								
25.	Mode of delivery	Spontaneous Vaginal Delivery (SVD) [ ]							
		Vaccum Extraction [ ]			Caesarean Section [ ]				

BABY

26.	Sex	1. Male [ ]	2. Female [ ]
27.	Birth Outcome	1. Live Birth [ ]	2. Still Birth [ ]

28.	Birth weight, kg		
29.	APGAR Score, 5 min		
30.	Congenital abnormalities	Yes [ ]	No [ ]

PART G: PLACENTA BLOOD MALARIA RESULTS

Light Microscopy results

Malaria Parasite Species and Stage	**Parasite density/µL Enter 000000 if not seen**					
P. falciparum asexual density, parasites/µL						
P. falciparum gametocyte density, parasites/µL						
P. malariae asexual density, parasites/µL						
P. malariae gametocyte density, parasites/µL						
P. ovale asexual density, parasites/µL						
P. ovale gametocyte density, parasites/µL						
P. vivax asexual density, parasites/µL						
P. vivax gametocyte density, parasites/µL						

Appendix C: Ethical Approval Letter