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Incidence of malaria parasites among pregnant women attending antenatal in Dutsin-Ma General Hospital, Katsina State, Nigeria

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Abstract

The study is to determine the incidence of malaria parasites among pregnant women attending antenatal in Dutsin-Ma general hospital, Katsina State between May-July 2021. Both thick and thin films were made and Giemsa stained using standard procedure provided by the NMEP/HJFMRI malarial diagnostic training, preparation and manual. Questionnaire was also distributed to the respondents. Overall, 98 (65.33%) of the 150 pregnant women investigated was found to have malaria infection in which the age group 21-30 had the highest frequency rate (67.90%). It was found that women in their 3rd trimester have high incidence rate of (78.57%), this result showed that respondents knew about Rapid Diagnostic test (52%), Microscopic (36%), Self-diagnostic (12%) and *Plasmodium* *falciparum* as dominant specie with 61 (40.67%) and *Plasmodium malaria* 37 (24.67%). In conclusion, the current study shows a very high incidence of malaria infection among pregnant woman attending ante-natal clinic in Dutsin-Ma general hospital due to high incidence of *P. falciparum* and respondents had good knowledge of malaria transmission by mosquitoes and prevention by the use of insecticide treatment nets. Following the high incidence of *P. falciparum* in pregnant women, more efforts are needed in the control of malaria in pregnancy, Regular environmental sanitation and antenatal booking for effective monitoring and prompt treatment of malaria in pregnancy will contribute significantly in reducing maternal morbidity and mortality cases.

Keywords: Incidence, Malaria, Parasite, Pregnant women, Antenatal, Hospital

Introduction

Malaria is a severe disease caused by the protozoan plasmodium that pass on to people through the bites of infected Anopheles mosquitoes, called "malaria vectors" which bite mostly between nightfall and dawn (Conroy *et al.*, 2012)^[8]. Plasmodium is a parasite accountable for causing malaria which infects the red blood cells and is spread by a female Anopheles mosquito (Conroy *et al.*, 2012)^[8]. It is a severe public health problem in tropical and subtropical regions of the world and caused by five Plasmodium species namely; *Plasmodium vivax, Plasmodium falciparum, Plasmodium ovale, Plasmodium malariae* and *Plasmodium knowlesi*. However, of these species; *P. falciparum* and *P. vivax* pose the greatest threat in the world (Conroy *et al.*, 2012)^[8]. The disease is transmitted to people mostly via the bites of infected female Anopheles mosquitoes (WHO, 2017)^[24]. *P. falciparum* infection is the commonest and the deadliest (Miller and Marley, 1999)^[17].

Malaria continues to be a cause of great suffering in tropical and sub-tropical regions of the world (Brabin, 1989)^[6]. Currently, it is endemic in about 100 countries affecting 4% of world's population (WHO, 2010)^[25]. Malaria has been eliminated or effectively suppressed in several parts of the world in the past decade but is now undergoing resurgence. It is returning to areas from which it had been eradicated as well as spreading into new areas such as Central Asia and Eastern Europe (Chrisantus, 2012)^[7].

Malaria increases vulnerability to other infections and slow down growth and development in children (Davidson, 2000)^[9]. It is associated with considerable economic burden including direct loss to government productive work or education (Davidson, 2000)^[9]. Malaria killed Northern Nigerian every 30 seconds; hence, pregnant women and their unborn children are also exposed to malaria which serves as major cause of maternal anemia and parental death (Davidson, 2000)^[9].

More than 40% of the world population lives in malaria prone areas. It is estimated by the world health organization that the number of malaria cases is 225 million in 2009 with about 781 000 deaths in 2009 (WHO, 2010)^[25]. Malaria is the most highly prevalent tropical disease with high morbidity and mortality, and with high economic and social impact (WHO, 2010)^[25]. Over 90% of all deaths caused by malaria occur in Sub - Saharan and tropical Africa (WHO, 2010)^[25]. In addition, pregnant women are at enormous risk of malaria due to natural immune depression in pregnancy (Fievet *et al.*, 2007). About 25% of all estimated malaria cases by the World Health Organization African Region occur in Nigeria (WHO, 2010)^[25]. The burden of malaria infection during pregnancy is caused mainly by *P. falciparum*, the most common malaria species in Africa (WHO, 2010)^[25]. Each year at least 3 million pregnancies occur among women in malarious areas of Africa, most of who reside

in areas of relatively stable malaria transmission (Brabin, 2000)^[5]. The symptoms and complications of malaria during pregnancy differ with the intensity of malaria transmission and thus with the level of immunity the pregnant woman has acquired (Perlmann and Troye-Blomberg, 2000)^[22].

In malaria endemic areas, majority of pregnant women may remain asymptomatic but still associated with complications on the mother and her foetus (Desai et al., 2007). They also serve as reservoirs and act as transmitters of infection. Malaria in pregnancy, especially when caused by P. falciparum, poses substantial risk to the mother and foetus by increasing the risk of foetal death, prematurity, low birth weight, and anaemia (Desai et al., 2007).

Many studies regarding prevalence of malarial parasites among pregnant women have been carried out in different hospitals. However, there is paucity of data in Dutsin-Ma general hospital concerning this problem. Therefore, this study aimed at determining the incidence of malarial parasite among pregnant women attending Dutsin-Ma general hospital.

Material and method Study area

This study was carried out in Dutsin-Ma town, Dutsin-Ma local government area of Katsina state, which lies between the coordinates of 12°27'17"N, 7°29'29"E (Neeru, 2015).

The local government has an area of 527 km² and a population of 169, 671 people as of 2006 census (Wikipedia, 2014). It is inhabited mostly by Hausa and Fulani speaking tribes and most of them are involved in farming and rearing of animals.

Sample size

One hundred and fifty (150) pregnant women attending the Antenatal clinic of General Hospital Dutsin-Ma Katsina state were randomly sampled by collecting their blood samples for analysis.

Selection criteria

The 150 pregnant women selected were women of reproductive age - between age 15-45 who were pregnant during the period of this research and attended the general hospital Dutsin-Ma, Dutsin-Ma L.G.A Katsina state.

Questionaire

A well-structured and pre-tested Questionnaires were designed to provide detailed information on demographic risk factors associated with malaria such as age, use of ITN, knowledge of malarial and diagnosis on each patient. An interview administered to the respondents in line with the criteria. The Questionnaires were translated into the local "HAUSA" language as majority of the respondents were deficient in English language.

Sample collection

Sample collection (For venous blood): to be obtained by venipuncture, Storage in anticoagulant: ethylen diamine tetra-acetic acid (EDTA) and another drop at the center of the slide. The right drop was spread immediately using the edge of a slide to make the thin smear, while the central drop was spread to make the thick smear which was covered about 15x15mm at the left side of the slide as recommended by WHO (2002)^[23].

Fixing and staining

Thick and thin blood films as described by Benishangul (1998)^[4] were made on clean slides and labeled accordingly as recommended by WHO (2002)^[23]. To prepare the thick film, 2 drops of blood sample were placed on a slide using a Pasteur pipette. The blood was gently spread for 20 seconds using the corner of a second slide to defibrinate the blood and to obtain a round smear. The slide was immersed in the staining trough, containing Giemsa solution prepared with buffered water in the ratio of 1:20, and contact was maintained for 30 minutes, and then allowed to dry.

For the thin film, a drop of blood sample was placed on a clean slide using a Pasteur pipette. The edge of a second slide was then laid on the drop of blood that would spread on the entire line of contact between the two slides. The second slide, steadily held to form a 45° angle with the original slide was then moved to the opposite end of the slide to which the drop was originally located. The thin film was fixed using Methanol (methyl alcohol) by maintaining contact with the methanol for 10 seconds. The slide was immersed in the staining trough containing Giemsa solution prepared with buffered water in the ratio of 1:10, and contact was maintained for 30 minutes, and then allowed to dry.

Microscopy

The thick films were used to determine the presence of malaria parasite and to determine the parasite densities while thin films were used to identify the parasite species and infective stages. Stained slides were examined under the light microscope using x40 and X100 objective lens (immersion oil).

Taking the number of leucocytes per microliter of blood as 6,000, parasite density of blood, this was expressed as: parasite count x 6,000 divided by number of white blood cells counted.

(Number of parasite counted \times 6000)/200 = parasite/ microliter of blood

Where 6000= average WBC count of individual 200= number of WBC counted.

Parasite detection and counting

The parasite was counted in thick film. The results were classified as scanty if less than 10 asexual forms of P. falciparum were seen per 100 thick field, (+) when 1-10 asexual forms of P. falciparum were seen per 100 thick fields, (++) when 11-100 parasites were seen per 100 thick fields and (+++) when 1-10 parasites were seen per thick field films. A blood slide was considered negative when the examination of 100 thick film fields did not show the presence of asexual forms of Plasmodium.

Ethical approval

The study was approved by the ministry of health Katsina MOH/ADM/SUB/1152/1/466 and the ethical committee of the General hospital Dutsin-Ma Katsina state. Informed consent was sought from the respondents and confidentiality of information given was maintained.

Data analysis

Data collected were analyzed using descriptive statistics and chi square (X^2) test. Tables were drawn to show the

differences and similarities between parameters. The variables and parameters selected for the study were analysed using statistical package for social science (SPSS) version 20.

Conflict of Interest

The authors declared no conflict of interest

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Results

Identification of Malaria Parasite from Blood samples of Pregnant Women attending Dutsin-Ma General Hospital Microscopic examination of Giemsa stained of thin and thick blood films was carried out at the general hospital Dutsin-Ma Katsina. Smears were prepared and slide was examined under x100 oil immersion objective of the microscope. The parasite is identified based on morphological characteristics of malaria parasite, the thick films was used for detection and counting of malaria parasite as shown in the table below.

Table 1: Microscopic Identification of Plasmodium Species	Table 1: Micros	scopic Identification	of <i>Plasmodium</i> Species
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S/N	Plasmodium Species	Morphological appearance under Microscope
1	Plasmo dium falainamum	Have delicate cytoplasm, small chromatin dot and regular cytoplasmic ring, some broken and comma shape
1 Plasmodium falciparum	with single or double chromatin dot	
2	Plasmodium ovale	Chromatin large and irregular in shape cytoplasm small and pigment scattered
3	Plasmodium vivax	Vivax have red chromatin dot and blue cytoplasm in schizont, chromatin divide into smaller irregular masses
4	Plasmodium malaria	Malariae have scattered pigment rings form fairly and chromatin dot at the center, usually fill the host.

Incidence of Malaria Parasites among the Study Population

The table below contains the incidence of plasmodium species among the study population. It can be seen that P. falciparum is the specie that has the highest incidence 61(40.67%) in the population surveyed followed by P. malariae which has 37(24.67%). The statistical analysis showed no significant difference ($x^2 = 137.933$; df = 3; p < 0.05) indicating that there is no significant relationship between the species.

Table 2: Incidence of Plasmodium Species among the study population

Species	No examined	No. of Positive Cases	Incidence (%)
Plasmodium falciparum	150	61	40.67
Plasmodium malariae	150	37	24.67
Plasmodium vivax	150	0	0
Plasmodium ovale	150	0	0

Incidence of Malaria by Trimester

In order to know the stage during pregnancy does the respondents treated malaria, their opinions were sought in this regard. The result is presented in table 3.

Based on the result of the analysis below, the pregnant women experienced malaria infection mostly in their 3rd

trimester (14, 11 - 78.57%) followed by 2^{nd} and 1^{st} trimesters (23, 17, 73.91). The least among them experienced malaria in their 1st trimester. The statistical analysis showed no significant difference ($x^2 = 19.582$, df = 3; p < 0.002) revealing there is no relationship between malaria infection and trimester.

Table 3: Incidence of Malaria by Trimester

Trimester	No. examined	No. of Positive Sample	Incidence (%)
1 st trimester	71	39	54.93
2 nd trimester	41	30	73.17
3rd trimester	14	11	78.57
2 nd and 3 rd trimesters	23	17	73.91
No Treatment	1	1	Total = 1.0

$$(X^2 = 19.582; df = 3; p = 0.002)$$

Presentation of the respondents' demographic characteristics

The demographic characteristics of the respondents selected along their opinion to determine the incidence of malarial parasites among pregnant women attending Dutsin-Ma general hospital were age, marital status and educational background. These variables were considered to have a bearing on the occurrence and spread of malarial parasite among pregnant women in the area selected for the study. Each of the variables is classified into frequencies and

percentages.

The table 4 shows that majority of the respondents are within the ages of 20 and 30 years (85%). It can also be seen that most of them holds a secondary school certificate (70%), while 26% of the respondents holds primary school certificate and only 4% attended tertiary institutions. It can be observed all the pregnant women are married (100%). This is possible because most women are typically married before getting pregnant.

 Table 4: Classification of the respondents by their demographic characteristics

Variable	Variable options	Frequency	Percent
A an Doman	20 - 30 years	85	85
Age Range	31 - 40 years	15	15
Marital Status	Single	0	0
Marital Status	Married	100	100
	Primary	26	26
Level of Education	Secondary	70	70
	Tertiary	4	4
	Total	100	100

Respondents' opinion on Incidence of Malaria

The opinion of the respondents was sought about the causes of malaria, signs and symptoms of the disease and their knowledge of malaria. This becomes imperative in order to effectively measure their understanding of the knowledge. Each of the variables was classified into frequencies and percentages in the table.

The table below shows that all the pregnant women surveyed for the research had knowledge of malaria. This was evident as majority of the respondents' suggested mosquitoes is the major causes of malaria (93%) with only 7% saying stagnant water. Fever was the commonest symptoms experienced by the pregnant women during their infection with malaria (50%), followed by headache (26%), body pains (8%), chill and shivering (4%), vomiting (3%) dizziness (2%) while only 1% suggested vomiting as the symptoms experienced.

Variable	Variable options	Frequency	Percent
Knowledge of malaria Causes of malaria Common Signs and Symptoms of Malaria	Yes	100	100
Knowledge of malaria	No	0	0
	Mosquitoes	93	93
Causas of moloria	Stagnant water	7	7
Causes of mataria	Dirty surroundings	0	0
	Yes100No0Mosquitoes93Stagnant water7Dirty surroundings0Don't know0Fever50Headache26Chill and Shivering4Body pains8	0	
	Fever	50	50
Common Signs and Symptoms of Malaria	Headache	26	26
	Chill and Shivering	4	4
	Body pains	8	8
	Loss of appetite	4	4
	Vomiting	3	3
	Weakness	1	1
	Dizziness	2	2
	Don't know	2	2
Total		100	100

Table 5: Classification of the Respondents opinion on Prevalence of Malaria

Respondents' opinion on prevention of malaria, malaria diagnosis and dissemination of malaria diagnosis information

In order to understand the prevalence of malaria among pregnant women in the area selected for the study, it is imperative to measure their understanding of malaria diagnosis, types of malaria diagnosis, its importance and the best way to disseminate information about malaria diagnosis. Their responses are presented in frequencies and percentages in the table.

It can be deduced from the table below that majority of the pregnant women surveyed for the study sleep under mosquito net as a prevention strategy against malaria. This is not surprising as majority possessed the knowledge of causes of malaria. It can also be seen that the respondents understand the knowledge of malaria diagnosis (86%) with

only 14% suggesting otherwise. The pregnant women also emphasized the importance of laboratory test before treatment of malaria as opined by majority (88%) with only 3% disregarding its importance.

Furthermore, it can be seen that Rapid Diagnostic Test is the commonest type of malarial diagnosis known to the respondents (52%) followed by Microscopy (36%) and Self Diagnosis with 12%.

Lastly, the respondents suggested that places of worship are the best way to disseminate information on malaria diagnosis (57%) as most of them tends to attends programs organized in the places. The respondents also suggested community meetings as a great way of sharing information about the disease while other suggested the use of pamphlets (4%) and 19% of them prefer other means such as radio, newspaper or word of mouth.

Table 6: Respondents Opinion on Prevention of Malaria, Malaria Diagnosis and Diagnosis	issemination of Malaria Diagnosis Information
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Variable	Variable options	Frequency	Percent
	Yes	72	72
Use of mosquito net	No	28	28
Knowledge of melonic discressio	Yes	86	86
Knowledge of mataria diagnosis	No	14	14
	Self-Diagnosis	12	12
Knowledge of malaria diagnosis Knowledge of types of malaria diagnosis Importance of laboratory test before treatment of malaria	Microscopy	36	36
	Rapid Diagnostic Test	52	52
	Very important	88	88
Importance of laboratory test before treatment of malaria	Barely important	9	9
	Not important	3	3
Dissemination of information on malaria diagnosis	Place of worship	57	57
Dissemination of information on malaria diagnosis	Pamphlets	4	4

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	Community meetings	19	19
	Others	20	20
Total		100	100

Discussion

Malarial disease is dangerous; especially on infection with P. falciparum is more hazardous during pregnancy. Pregnancy appears to interfere with the immune processes in malaria, a disease which itself alters immune reactivity. The incidence of clinical malaria is higher and severity greater in pregnant women than non-pregnant women (Okwa et al., 2003)^[3]. In this study, the incidence rate of *P. falciparum* in pregnant women was 40.67% which is in line with 55.9% reported by Aribodor et al. (2012) and 72% by O.A Adefioye et al. (2007)^[19] respectively. Maestre et al. (2014) further explain that, apart from P. falciparum, infections by P. vivax during pregnancy leads to rare mortality associated with multiple relapses. However, the findings of this study on the incidence of *P. falciparum* are higher than the report of Aina et al. (2018)^[3] who reported 2% incidence. In this current study incidence of 65.33% malaria parasite was recorded which is in agreement with the research of Abdullahi et al. (2020)^[1] and Omang et al. (2020)^[20] who reported 72% and 79% respectively. Though, the incidence of this study is lower than these two mentioned above. The differences may be due to the nature of Ante-natal clinic about malarial control strategies like use of insecticide treated nets (ITNs). Younger women appeared to be susceptible to malaria in this study as incidence was highest among age group of 21-30 (67.90%). This is closely related to the study of Aina et al. (2018) [3] with 46.5% in 25-30 years age group but contradict the findings of O.A Adefiove et al. (2007)^[19] with 72% among 36-40 years of age and that of Maureen et al. (2016)^[16] who reported 36-40 years old group to be susceptible. Moreover, the study agreed with Aina et al. (2018)^[3] who stated that adolescent and young adult pregnant women were more susceptible to malaria than older pregnant women because of continuous development of malaria immunity in older women.

In this study 3rd trimester was found with the highest incidence 78.57% then 2nd and 1rd trimester with incidence of 73.91% and the least was 1st trimester with the incidence of 54.93%. This is in line with the finding of Maureen et al. (2016)^[16] and Abdullahi et al. (2020)^[1] who reported the highest incidence in 2nd trimester and least incidence in 1st trimester but contradict with Conroy et al. (2012)^[8] with the highest incidence in 1st trimester followed by 3rd trimester and the least was 2nd trimester. It was observed that women in their 1st trimester had higher incidence according to previous study. However, these findings reported that over 70% of the infections were in the 3rd trimester. In contrast to other findings, observed that 1st trimester is lesser during pregnancy. The reason may be due to negligence in Antenatal during early pregnancy and most of them may have unrecognized and untreated malaria infection as most infection is asymptomatic or peak incidence of plasmodium species (Maureen et al., 2016)^[16].

It was imperative to discover that sign and symptoms of malaria is relatively high with fever (50%), Headache (26%), Body pains (8%), Vomiting (3%) reported by Abdullahi *et al.* (2020) ^[1]. This is in line with the current study which respondents indicated that malaria can be caused by Mosquito bite with 93%, stagnant water 7% and 100% of the respondents were having the knowledge of

malaria. Most of the respondents acknowledged the importance of diagnosis with 88%. Based on the prevention in this study and those that mentioned sleeping under insecticide treated nets (ITNs) were 72% This is in agreement with 113(56%) reported by Aina *et al.* (2018)^[3]. The incidence of malaria parasite among the plasmodium species of the population was 61(40.67%) of *P.* falciparum which arose as the dominant parasite among pregnant women attending Dutsin-Ma general hospital. This correlate with the study of Meastre *et al.* (2014) and Aina *et al.*, (2018)^[3] followed by *P. malariae* with 37(24.67%). In this study, the statistical analysis showed no significance difference between the malarial parasites (x² =137.933; df=3; p< 0.05).

Conclusions

The current study shows a very high incidence of malaria infection among pregnant women attending ante-natal clinic in Dutsin-Ma general hospital with 65.33%, and respondents had good knowledge of malaria 100%, transmission by mosquitoes 93%, stagnant water 7%, sign and symptoms; Fever 50%, Headache 26%, and prevention by the use of insecticides treated nets 86%. Also, by age group, 20-30 years had the highest incidence of malaria with 85%.

Therefore; based on the study, the high incidence of malaria parasites among pregnant women attending ante-natal in Dutsin-Ma general hospital could be due to rainy season, poor environmental sanitation and stagnant water favored the breeding of mosquitoes in the study community.

Recommendations

Microscopic examination proved to be more effective in detection of low parasite density; as such, microscopic examination should be widely employed in health institutions and laboratory for adequate diagnosis and detection of malaria parasite.

Governmental and Non-governmental organization should be encouraged to carry out awareness campaign on the use of ITNs, Environmental hygiene, as well as preventive drugs to reduce the incidence in the study area.

Also, regular environmental sanitation dislodge mosquitoes from their breeding places and early Ante-natal booking for effective monitoring and prompt treatment of malaria in pregnancy will contribute significantly in reducing maternal morbidity, mortality and perinatal mortality.

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