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Impact of *Plasmodium falciparum* Parasitaemia on Platelet Levels among Pregnant Women Attending State Specialist Hospital Maiduguri, Borno State Nigeria

B. Timothy¹, Y. Inuwa¹, M.A. Muhammad¹, A.B. Suleiman¹, J.Y. Dahiru², K.A. Makinta¹

¹Department of Zoology, Faculty of Life Sciences, University of Maiduguri, Maiduguri, Nigeria.

²Department of Biological Sciences, Faculty of Life Sciences, Federal University of Kashere, Nigeria.

*Corresponding author's Email: mamuhammad@unimaid.edu.ng, doi.org/10.55639/607.02010042

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ABSTRACT

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Malaria is a major public health problem in Sub-Saharan Africa. Most of the infections are asymptomatic, affecting mainly pregnant women and children. This was conducted to investigate the impact of *P. falciparum* parasitaemia on Platelets amongst pregnant women attending State specialist hospital Maiduguri, Borno State, Nigeria. Blood samples were collected from 384 pregnant women and subjected to microscopy for detection of malaria parasite using thick and thin blood films, whereas the parasite density was counted by the method of WHO, 2008 while the Platelets was estimated using sysmexhaematology auto-analyser. A total number of 304 (79.16%) tested positive of *Plasmodium falciparum* malaria, and 80(20.83%) tested negative of *Plasmodium falciparum* malaria. It was observed that there was a negative correlation between the parasite densities and Platelets among the infected pregnant women ($r=-0.9304$, $P=0.05$) as well as between parasite density and trimesters ($r=-0.949$, $p=0.05$), ($r=-0.9179$, $P=0.05$), and ($r=-0.943$, $P=0.05$) respectively. In conclusion, Malaria infection can affect some haematological parameters of the infected pregnant women and therefore regular and consistent screening of malaria should be conducted on each hospital visits.

Corresponding author: M.A. Muhammad, Email: mamuhammad@unimaid.edu.ng
Department of Zoology, Faculty of Life Sciences, University of Maiduguri, Nigeria

INTRODUCTION

Malaria is an acute febrile illness caused by *Plasmodium* species, which are spread to people through the bites of infected female *Anopheles* Mosquitoes. Malaria remains one of the major health challenges worldwide including Nigeria, it has become a major cause of morbidity and mortality particularly among pregnant women and children under age of five (Jensen *et al.*, 2017; Inuwa *et al.*, 2021). *Plasmodium falciparum* is the deadliest *Plasmodium* species and the most prevalent on the African continent. Malaria is a parasitic disease caused by *Plasmodium* species, in the phylum Apicomplexa (WHO, 2020).

In 2020, *Plasmodium falciparum* infection during pregnancy increases the risk of spontaneous abortion still birth, preterm delivery, low birthweight and maternal death globally, over 120m pregnant women were at risk of *Plasmodium falciparum* (Muhammad *et al.*, 2025).

It was reported to be a common cause of miscarriage in pregnant women, premature delivery, low birth weight, maternal anemia, intrauterine growth retardation and intrauterine death (Aduloju *et al.*, 2013). Malaria during pregnancy is therefore a serious case in sub-Saharan Africa, affecting an estimated 24 million pregnancy women annually. Although *Plasmodium falciparum* infection in pregnancy is usually asymptomatic, it often contributes to adverse perinatal outcomes with a high risk for infant death, particularly in areas of lower malaria endemicity. Pregnancies in women living in malaria endemic regions particularly Africa are associated with a high frequency and density of *Plasmodium falciparum* Parasitemia, with high rates of maternal morbidity, including fever and severe anemia, abortion. Each year, between 75,000 and 200,000 infant deaths are attributed to malaria infection in Africa (Muhammad *et al.*, 2025).

Malaria has been shown in many studies to worsen certain pregnancy outcomes (Rich *et al.*, 2009). These include an increased incidence of anaemia and spontaneous abortions. Others include intrauterine growth restriction (IUGR), stillbirths, prematurity, low birth weight, fetal distress and congenital malaria (Asa and Onayade, 2008). The biological basis for these adverse outcomes has been extensively studied. Erythrocytes infected with *Plasmodium falciparum* accumulate in the placental bed. This is through adhesion of the infected erythrocytes to molecules of Chondroitin a present in the placenta (Perlman and Troye, 2000).

A prevalence of placental parasitaemia of between 10 and 45% in malaria endemic areas has been reported with significant *Plasmodium falciparum* dominance

(Bruce, 1987). Pregnancy increases the frequency and severity of most infectious diseases but its effect on malaria seems worse (WHO, 2019).

Haematological changes are some of the most common complications in malaria and they play a major role in malaria pathogenesis. These changes involve the major cell types such as RBCs, leucocytes and Platelets (Ballem, 2018). Malaria infected patients tended to have significantly lower platelets, WBCs, lymphocytes, eosinophils, RBCs and Hb level, while monocyte and neutrophil counts were significantly higher in comparison to non- malaria infected patients (Jensen *et al.*, 2017). One study showed patients with higher WBCs count compared with community controls, the most common complication during malaria infection is thrombocytopenia (Sullivan and Martin, 2015).

The present study was aimed to examine the Impact of *Plasmodium falciparum* Parasitemia on Platelets among Pregnant Women Attending State Specialist Hospital, Maiduguri.

MATERIALS AND METHODS

Study Area

Maiduguri Lies on latitude 11° 40'N and longitude 13° 05'E. The state occupies the greater part of the Chad basin and is in the Northeastern part of Nigeria, the state share borders with the republic of Niger to the North, Chad to the Northeast and Cameroon to the East. Within Nigeria, the state shares boundaries with Adamawa state to the south, Gombe state to the west and Yobe state to the Northwest. Maiduguri is the Capital of Borno State. It is located in the Sahel Savannah region of northeast Nigeria. The climate of Maiduguri is favorable, with a mean annual rainfall and temperature of about 650 mm and 32°C respectively. The month of March and April are the hottest periods of the year with temperatures ranging between 30°C and 40°C. It is usually cold and dry during the harmattan, November to January being the coldest months. (Muhammad *et al.*, 2020). Borno state has an area of 69, 435 square kilometers about 7.69% of the total land area of the country. The 2006 census figure has the state population of 4,151,193 with population density of approximately 60 inhabitants per square to meter (NPC, 2006).

Ethical Clearance and Consent to Participate

Ethical approval for the study was obtained from the Chairman Ethical Committee of the State Specialist Hospital Maiduguri, Borno State (Number: SSH/ADM/621/VOL.1/127). Confidentiality of the participants and the information provided was assured and maintained throughout the study period.

Study Population and Sample Size Determination

Three hundred and eighty-four (384) blood samples were collected from pregnant women attending antenatal at State Specialist Hospital Maiduguri, Borno State between August and September. Sample size was determined using the following formula by Muhammad *et al.*, 2020.

Where z = standard normal deviation and probability at 95% confidence level (1.96), p = prevalence or proportion of value to be estimated from previous studies

q = proportion of failure (= $1-P$)

d = precision, tolerance limit

$n = z^2 pq / d^2 = 1.96^2 \times 0.5 \times 0.5 / 0.05^2 = 384$

Hence, A total Of 384 samples

Inclusion Criteria and Exclusion Criteria

Consented Pregnant women receiving antenatal care at the specialist hospital Maiduguri Borno State participated in the study. Exclusion criteria were participants with severe illness and in need of immediate medical attention, and those that did not give their consent were excluded in this study.

Preparation and Microscopy Examination of blood film

Each pregnant woman that participated in this study was given a unique identification number and the numbers were labeled on individual EDTA-containers. Venipuncture was used to obtain blood samples from the pregnant women into a labeled EDTA (Ethylenediaminetetraacetic acid)-containing tube. Blood samples were taken and analyzed on the same day they were taken. A drop of the pregnant women's blood was taken from the EDTA container using a micropipette and was spread out on a labeled, clean and grease free microscope slide to make a thick blood smear. The prepared slides were allowed to dry and then stained with Giemsa stain to give the parasite a distinguishing appearance before examining under an x100 oil immersion objective of the light microscope. The entire bulky film was scanned in its entirety. When no malaria parasite is seen on a thick film, it was considered negative. Where parasites are found, the rest of the blood film were inspected to eliminate the potential of a mixed infection being missed. All of the species and stages that were observed were identified and documented. *Plasmodium falciparum* was the most common *Plasmodium* species discovered in this investigation. They were discovered in trophozoites and ring forms.

Preparation of thick blood film (WHO, 2000)

A drop of each blood sample was placed on the center of a clean grease-free microscope glass slide. An applicator was used to spread the blood in small circular forms. The slides were kept to air dry. The

blood films were dipped into field stain A (eosin) for 5 seconds. It was rinsed off gently in clean water. The slides were then allowed to air dry (WHO, 2000)

Preparation of thin blood film (WHO, 2000)

A drop of each blood sample was placed on the center of a clean grease free microscope glass slide using a pasture pipette. A cover slip was placed on the drop of blood allowing blood to spread along the edge of the coverslip. The coverslip was then held at a suitable angle of 45° and pushed along the slide, drawing blood behind it, until the whole blood was smeared. It was then fixed briefly in methanol and allowed to dry. The thin film was placed over the staining rack and Giemsa stain poured over it. This was diluted and allowed to stand for about 10-20 minutes, before being rinsed with a large quantity of water. Underneath the slide was cleaned with cotton wool and placed on a rack to air dry (WHO, 2000).

Determination of Parasite Density

The thick film slide was stained for 10 to 15 minutes with 10% Giemsa for the assessment of parasite density. The samples were examined using objectives of a research microscope (x100) asexual parasites was counted alongside with 200 Platelets. In an event that parasite count was <10 parasites/200 leukocytes; count was continued per 500 Platelets. The parasite density was expressed as the number of asexual parasites per ml of blood by assuming a mean normal Platelets count of 8000/ μ l of blood Gilles and Warrell, 1993 and modified by (WHO, 2008).

Parasitaemia (per μ l) = number of parasites \times 8000 / number of White Blood Cell (200/500) as described by WHO (2008).

Blood Analysis

The collected blood samples were transferred to the laboratory for the estimation of blood parameter such as platelets by using sysmex hematology Auto analyzer, (2011). The result was recorded alongside findings of each subject's data.

The Standard Operating Procedure for Sysmex Hematology Auto analyzer (2011).

- (I) Switch on the machine and wait until, the visual Display unit (VDU) is in the "ready" mode, this takes about five (5) minutes (SOP, 2011).
- (II) Place the sample on a mixer to mix for about five (5) minutes.
- (III) Open the well-mixed sample of blood in EDTA and prime it with probe.
- (IV) Using the sysmex auto analyzer aspirate the sample to analyze automatically.
- (V) Remove from the auto-analyzer as bit sound is made.

- (VI) The result of blood parameters such as, Platelets and percentage are recorded.
- (VII) The result is recorded in result sheets after quality officer is certified.

Statistical Analysis

Data were entered into a computer and analysed using SPSS version 25.0 for Windows (SPSS Inc. Chicago, IL: USA). Simple percentages, Standard Deviation, Range and description was used for the analysis. Regression analysis was employed to assess the relationship between parasite densities and Platelets. Differences were considered significance ($P < 0.05$).

RESULTS

The data presented in Table 1, shows the results of the study on pregnant women attending State Specialist Hospital Maiduguri, Borno State. A total number of 384 pregnant women were enrolled for this research, 304 (79.16%) were tested positive, where 80 (20.83%) were tested negative. Mean and S.D was calculated to assess variability in the observed data set, as a result the age of the participants was considerably spread between 18-38 years from the mean \pm SD 282.99 ± 18.25 tested positive and 102.06 ± 11.03 tested negative. The table also indicates that the infection of *plasmodium falciparum* is more prevalence among pregnant women who were in their second trimester with prevalence rate (44.71%) compared to 1st trimester (9.37%) and third trimester (25%).

Generally, a highly significant association between malaria parasite density and Age of the subjects was observed, whereby 1000- 1500 was generally low or absent across all age categories. However, pregnant women within the age range 25-34 years had the highest prevalence of (27.6%; 84/304), ($X^2=19.67$, $P=0.001$) with regards to parasites density count greater than 500, as presented in Table 2.

Table 3 shows significant association between malaria parasitaemia and trimester of the subjects. In which pregnant women in their second trimester 84(27.6%), 85(27.9%), 12(3.9%) recorded the occurrence of parasite density counts of ≤ 500 (mild), 501-

1000(severe) and 1001-1500(chronic) respectively as compared to first and third trimester, which recorded the occurrence of PDC ≤ 500 (8.5%), 501-1000 (3.9%), 1001-1500 (0.00%) and ≤ 500 (15.7%), 501-1000 (11.8%), 1001-1500 (0.3%), ($X^2=11.02$, $P=0.003$) for first and third trimester respectively.

Table 4: The result of this study revealed a non-significant association between Platelet Counts and Age of the subjects in the study area. In which pregnant women between the age group 25-34 years recorded high number of malaria infected patients with low 118(38.8%), Normal 23(7.6%), and High 8(2.6%) ($X^2=4.69$, $P=0.320$) platelets counts.

Table 5: Revealed a highly significant association between platelet counts and trimesters of the pregnant women in the study area. In which the pregnant women in their 1st Trimester 124 (40.8%) recorded a high number of thrombocytopenia, while pregnant women in 2nd Trimester 24(7.9%), 10(3.3%), ($X^2=15.75$, $P= 0.003$) had high number of normal platelets counts and thrombocytosis.

Figure 1 shows the influence of parasites density count of malaria and pregnant women ($r^2= -0.9304$, $P=0.05$) in all trimesters and was considered highly significant which means the *Plasmodium falciparum* has a Negative impact on platelets in pregnant women infected with malaria. Furthermore, a highly significant association was also observed between parasite density and first trimester ($r^2= -0.949$, $p=0.05$), second trimester ($r^2= -0.9179$, $P=0.05$), as well as third trimester ($r^2= -0.9431$, $p=0.05$) as shown in Figure 2, 3 and 4 respectively. Figure 3&4 also indicates the relationship between PDC and platelets among pregnant women in second and third trimester respectively.

Figure 5 shows the relationship between platelets and parasites density in malaria infected pregnant women according to trimester shows that subjects in the third trimester recorded highest percentage (5.3%) of thrombocytopenia, and parasites densities and platelets have a Negative correlation.

Table 1: Characteristics Baseline of the Enrollment of the pregnant women attending State Specialist Hospital Maiduguri

Variables	Positive (%)	Negative (%)	Total (%)
No. Enrolled	304 (79.16%)	80 (20.83%)	384 (100%)
Mean \pm SD	282.99 ± 18.25	102.06 ± 11.03	385.05 ± 29.28
Range	18-39 years	18-39 years	18-39 years
TRIMESTERS			
1 st Trimester	36 (9.37%)	5 (1.30%)	41 (10.67%)
2 nd Trimester	172 (44.79%)	48 (12.50%)	220 (57.29%)
3 rd Trimester	96 (25.00%)	27 (7.03%)	123 (32.03%)

Table 2: Age- Based Distribution of Malaria Parasite Infection and Parasite Density among Positive Cases

Parasitic density of parasite/ μ L of blood	18-24years	25-34 years	35-39 years	Total	X ² – Value	P- Value
≤ 500	53(17.4%)	66(21.7%)	32(10.5%)	151(49.6%)	19.67	0.001
500-1000	40(13.1%)	84(27.6%)	20(6.5%)	144(47.3%)		
1000-1500	8(2.6%)	1(0.3%)	0(0.00%)	9(2.9%)		
Total	101 (33.2%)	151(49.6%)	52(17.1%)	304(100%)		

Table 3: Distribution of Malarial Parasite Density in-relation to Trimester among Positive Subjects in the Study Area

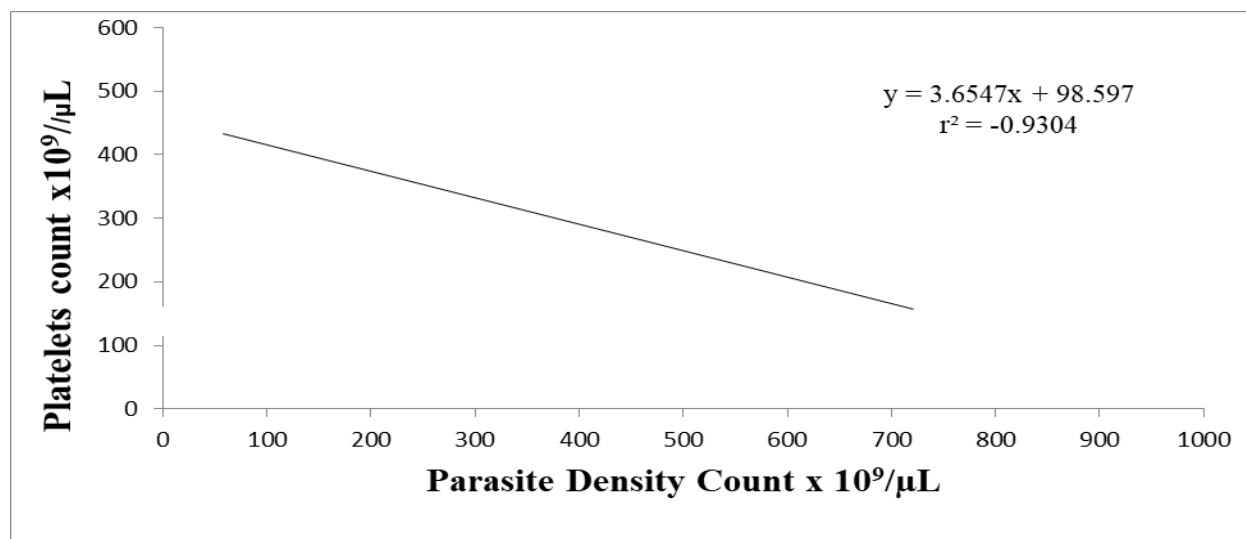
Parasitic density (no of parasite/ μ L of blood)	First trimester	Second trimester	Third trimester	Total	X ² – Value	P- Value
≤ 500	26(8.5%)	84(27.6%)	48(15.7%)	158(51.9%)	11.02	0.003
500-1000	12(3.9%)	85(27.9%)	36(11.8%)	133(43.7%)		
1000-1500	0(0.00%)	12(3.9%)	1(0.3%)	13(4.2%)		
Total	38 (12.5%)	181(59.5%)	85(27.9%)	304(100%)		

Table 4: Distribution of Platelet Counts in Relation to Age of the Subjects in the Study Area

Age (Years)	Low	Normal	High	Total	X ² - Value	P-Value
18-24	79(26.0%)	18 (5.9%)	3 (1.0%)	100(32.9%)	4.69	0,320
25-34	118(38.8%)	23 (7.6%)	8(2.6%)	149(49.0%)		
35-39	39(12.8%)	10(3.3%)	6(2.0%)	55(18.1%)		
Total	236 (77.6%)	51(16.8%)	17(5.6%)	304(100%)		

Table 5: Distribution of Platelet Counts in Relation to Trimester of the Subjects in the Study Area

Trimester	Low	Normal	High	Total	X ² - P-Value Value
1 st Trimester	124(40.8%)	17 (5.6%)	2 (0.7%)	143(47.0%)	15.75 0.003
2 nd Trimester	72(23.7.8%)	24 (7.9%)	10(3.3%)	106(34.8%)	
3 rd Trimester	40(13.2%)	10 (3.3%)	5(1.6%)	56(18.4%)	
Total	236 (77.6%)	51(16.8%)	17(5.6%)	304(100%)	

**Figure 1:** influence of parasite density (*P. falciparum*) on Platelets among pregnant women in all trimesters

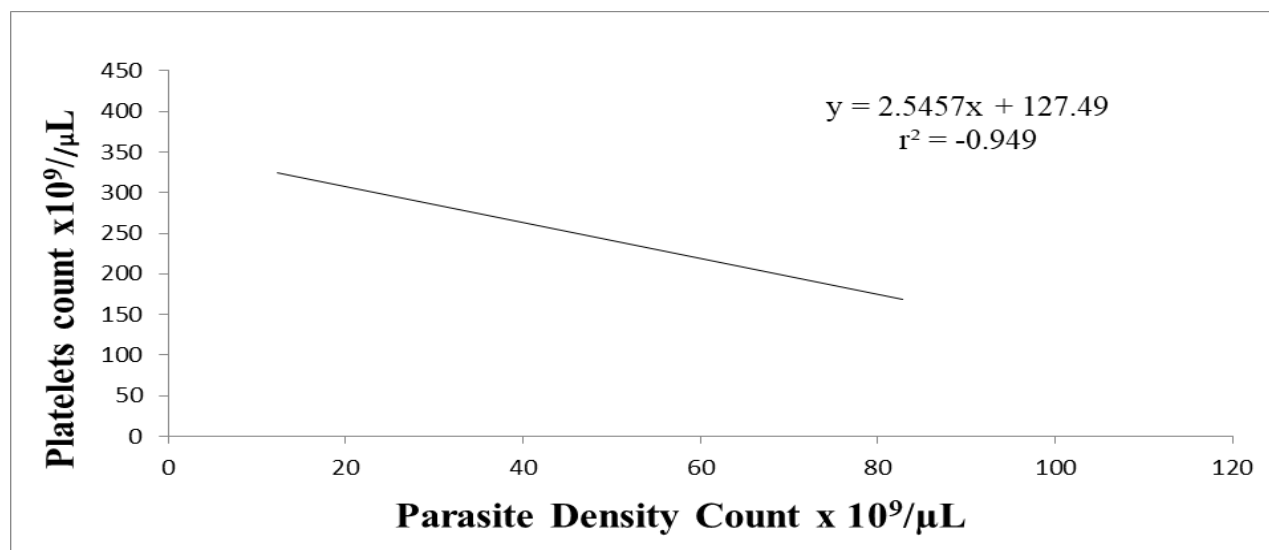


Figure 2: The influence of parasite densities (*P. falciparum*) on Platelets among pregnant women in first trimester

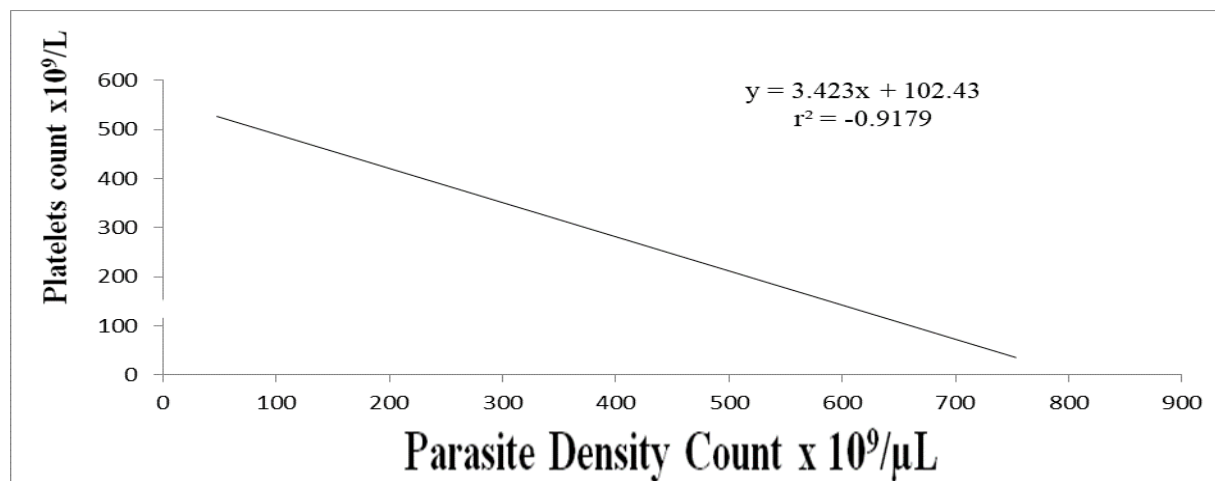


Figure 3: Influence of Parasite Density (*P. falciparum*) on Platelets among Pregnant Women in Second Trimester

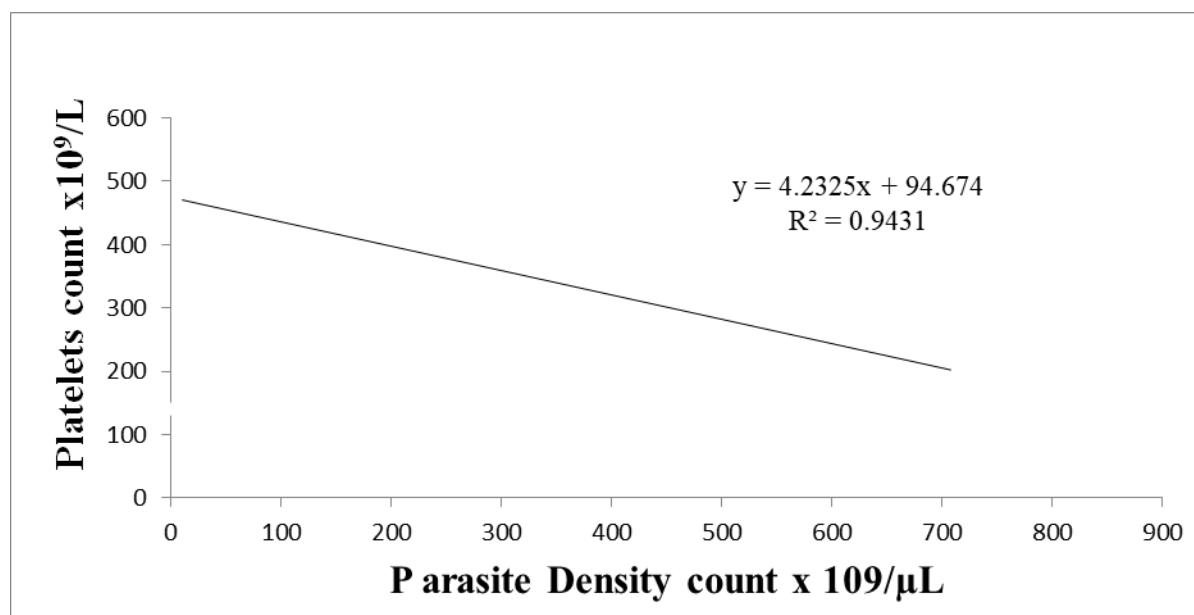


Figure 4: Influence of Parasite Density (*P. falciparum*) on Platelets among Pregnant Women in Third Trimester

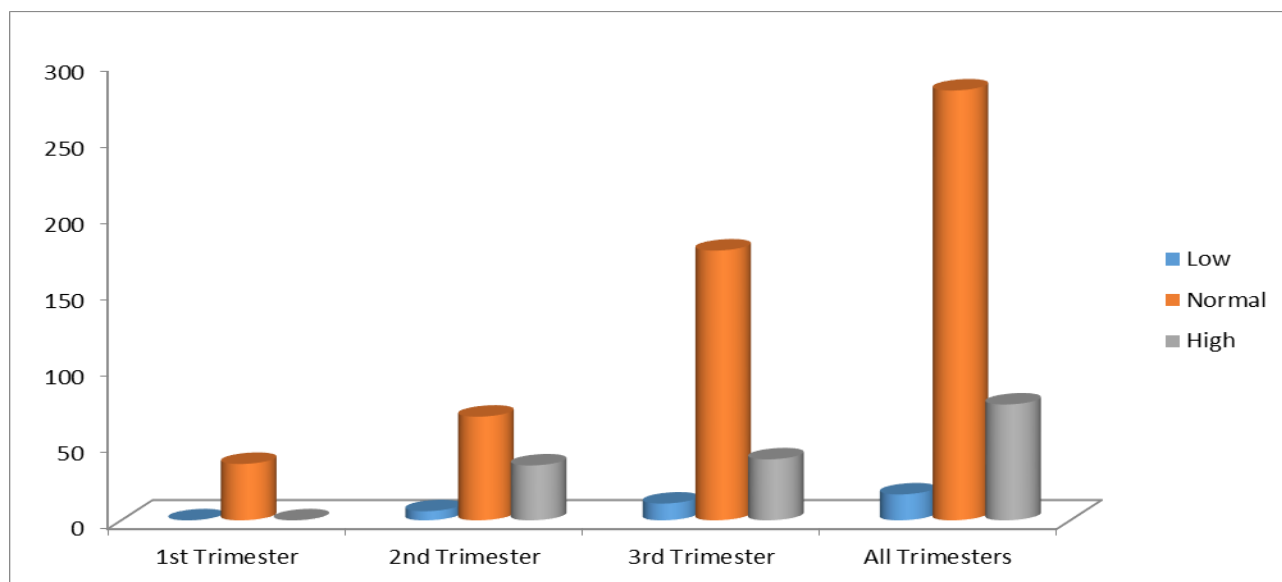


Figure 5: Relationships between Platelets and Parasite Density among Pregnant Women in All Trimesters

DISCUSSION

Haematological parameters are used as indices to monitor the severity of malaria, the degree of changes in haematological parameters depend on the level of parasitemia, nutritional status, malaria immunity, and the endemicity of the disease (Maina *et al.*, 2010).

The prevalence of *Plasmodium falciparum* malaria among pregnant women attending the State Specialist hospital, Maiduguri was 79.16% which is relatively high. This study is similar with the earlier study carried out in Southern part of Nigeria (77.9%) by Akingbola *et al.* (2018). But contradicted Mvondo *et al.* (2014) obtained low prevalence of malaria in pregnancy of 45.0%. The high prevalence recorded in this study area may be due to the fact that Maiduguri is malaria endemic area, and the climate and vegetation provides a breeding sites for the survival of mosquito larvae, therefore contribute to this prevalence. Similarly, the high seasonal rainfall gives rise to much surface water to support the breeding of vectors (Inuwa *et al.*, 2021). As the study was carried out from August to September (wet season) when infection rates was high due to stagnant water which accumulates due to poor drainage systems in the environment thereby favoring the reproduction of mosquitoes that act as vectors for malaria parasites and enhancing the incidence of the *Plasmodium* species. High rate of malaria transmission during rainy season has been reported by Muhammad *et al.* (2025).

The result of this study also reported a high prevalence of malaria among 2nd trimester (44.79%) as compared with other trimester counterpart. This study corresponds with earlier findings of Jensen *et al.*

(2017) who conducted a similar study in Ebonyi state. Our findings revealed low parasites density counts in 3rd trimester (25.0%). This disagrees with Ingrid *et al.* (2012) who observed the recovery in the 3rd trimester due to the satisfactory immune response to malaria infection, re-acquiring their pre pregnancy immune status at about the time of delivery. The observation could also be as a result of constant intermittent preventive treatment (IPT) given to these women during antenatal care visit which usually commence during second trimester (WHO, 2020).

The results of this study shows that pregnant women in their second trimester 84(27.6%), 85(27.9%), 12(3.9%) recorded high occurrence of parasite density counts of ≤ 500 (mild), 501-1000(severe) and 1001-1500(chronic) respectively as compared to first and third trimester, which recorded the occurrence of Parasite Density Counts ≤ 500 (8.5%), 501-1000 (3.9%), 1001-1500 (0.00%) and ≤ 500 (15.7%), 501-1000 (11.8%), 1001-1500 (0.3%), for first and third trimester respectively. This study contradicts Maina *et al.* (2010), who reported that pregnant women in their third trimester, had high parasite density counts.

Generally, parasite density counts 1000- 1500 was generally low or absent across all age categories. However, pregnant women within the age range 25-34 years had the highest prevalence of (27.6%; 84/304) with regards to parasites density count greater than 500, as presented in table 2. This study tallies with an earlier study of McCrae (2013), who observed that Severe malaria cases, especially those caused by *Plasmodium falciparum*, can lead to thrombocytopenia (low levels of platelets), malaria can disrupt the

normal balance of clotting and deficiency which can lead to increased bleeding risk.

The result of Table 4 revealed a non-significant association between Platelet Counts and Age of the subjects in the study area. In which pregnant women between the age group 25-34 years recorded high number of malaria infected patients with low 118(38.8%), Normal 23(7.6%), and High 8(2.6%) ($X^2=4.69$, $P=0.320$) platelets counts. This is consistent with McCrae, 2013 which reported thrombocytopenia in pregnancy. Moreover, our findings revealed a highly significant association between platelet counts and trimesters of the pregnant women in the study area. In which the pregnant women in their 1st Trimester 124 (40.8%) (Table 5) recorded a high number of thrombocytopenia, while pregnant women in 2nd Trimester 24(7.9%), 10(3.3%), ($X^2=15.75$, $P=0.003$) had high number of normal platelets counts and thrombocytosis. The results of this study tallies with the findings of Sullivan and Martin, 2015 which reported thrombocytopenia in 1st trimester of the pregnant women infected with malaria.

Our findings show a negative relationship between parasite density count and platelets of the pregnant women on 1st trimester. Also, it revealed a significant negative association between PDC and thrombocyte of the pregnant women in-p their 3rd trimester which agrees with the findings with the findings of Sullivan and Martin 2015. However, our study is limited to the effects of Malaria on platelets levels, this could be due to financial constraints and we therefore recommend further research on impact of malaria and all other hematological parameters such as White Blood Cell, Red Blood Cell, Hemoglobin etc., as this study focuses on platelets

Hematological changes are most common complication in malaria infection that plays a major role in malaria pathology which affect the major cell lines such as Platelets, red blood cells and leukocytes (Maina *et al.*, 2020). Thrombocytopenia develops in 5% to 10% of women during pregnancy or in the immediate postpartum period (Douglas, 2017). In this current study 5.20% thrombocytopenia was recorded among pregnant women, in their second trimester were recorded 3.12% and the results also indicates the strong relationship between parasite densities and platelets count. This study is not concurrent with the study conducted by Akingbola *et al.* (2018), who reported that thrombocytopenia is not caused by malaria. Due to hemodilution secondary to expansion of plasma volume, the Platelets count in normal pregnancies may decrease by approximately 10%, with most of this decrease occurring during the third trimester (McCrae, 2013; Ballem, 2018) although the

absolute Platelets count tends to remain within the normal reference range in most patients. (Burrows *et al.*, 2013; Sullivan and Martin 2015) After anemia, thrombocytopenia is the second most common hematologic abnormality that occurs during pregnancy.

CONCLUSION AND RECOMMENDATION

Malaria infection during pregnancy have adverse effects on maternal hematological parameters, particularly platelets, it is well established that anemia is the most common consequence of *P. falciparum* malaria infection. Malaria infection can increase some of the hematological changes associated with pregnancy and should be prevented to save lives of the mothers and the babies.

The high prevalence rate of malaria in pregnancy found in this study indicates that there is a need for the Borno state government to continuous ensure the implementation of malaria control and prevention activities in the state. Also, in stable malaria transmission areas such as Maiduguri, the ministry of health should ensure that all the healthcare facilities where pregnant women receive ANC implements the WHO's three evidence-based strategies for malaria prevention and treatment in pregnancy, that is; encouraging routine use of insecticide –treated nets, sealing up of intermittent preventive treatment of malaria in pregnancy with sulfadoxine-pyrimethamine at the second and third trimesters of pregnancy, and appropriate malaria case management through prompt and effective diagnosis and treatment of malaria in pregnant women.

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