International Journal of Innovative Research in Medical Science (IJIRMS)
Volume 07, Issue 05, May 2022,
https://doi.org/10.23958/ijirms/vol07-i05/1404

Original article

Congenital and Placenta Malaria and Newborn Birth Weight in Asymptomatic HIV Positive Pregnant Women at Term

Taiwo O. Kuku-Kuye [*1,2], Oluwaseun O. Orekoya [1], Adetokunbo O. Fabamwo [1,2], Oluwarotimi I. Akinola [1,2], Ayokunle M Olumodeji [1]

[1]Department of Obstetrics and Gynaecology, Lagos State University Teaching Hospital, Lagos
[2]Department of Obstetrics and Gynaecology, Lagos State University College of Medicine, Lagos

*Corresponding Author: Taiwo O. KUYE-KUKU; Orcid Id: *https://orcid.org/0000-0002-1721-8968;*
Email id: bethkuku@yahoo.co.uk

Received 30 April 2022; Accepted 20 May 2022; Published 30 May 2022

Abstract

Background: Human Immuno-deficiency Virus (HIV) and Malaria infection independently are major contributors to adverse perinatal outcomes in sub-Saharan Africa. A co-infection of both in pregnancy has been associated with more deleterious birth weight- related neonatal outcomes. We assessed for congenital malaria and evaluated the birth weights of babies born to HIV positive mothers with and without evidence of placenta malaria at term delivery. ***Methods:*** This was a comparative cross-sectional study in which 90 low-risk, HIV positive pregnant women on highly active retroviral therapy (HAART), asymptomatic for malaria, with uncomplicated delivery, at the Lagos State University Teaching Hospital, were consecutively recruited to have their maternal venous, placenta and newborn blood samples assessed following delivery for malaria. A placenta biopsy was also done to assess for features of placenta malaria. Data obtained were expressed in frequencies and percentages and variables were tested as appropriate using an independent t-test, chi-square and spearman's correlation. The level of significance was set at $p<0.05$. ***Findings:*** The prevalence of placenta malaria and congenital malaria in the study was 11.1% and 0.0% respectively. Only 3.3% of the asymptomatic HIV positive, pregnant women at delivery had positive malaria tests of their venous blood samples. The mean neonatal packed cell volume at birth for women with and without placenta malaria was 43.6% and 44.9% respectively ($p=0.044$). The mean birth weight of the babies born to HIV positive women with and without placenta malaria was 2.7±0.2kg and 3.0±0.4kg respectively ($p=0.008$). The correlation between birth weight and CD4 count was positive but insignificant ($r=0.069$, $p=0.515$). ***Conclusion:*** Although, irrespective of placenta malaria status, congenital malaria is a rarity in babies of HIV positive pregnant women, the birth weight and packed cell volume of babies born to HIV positive women is significantly lower in this group of women with features of placenta malaria than in women without placenta malaria.

Keywords: placenta, malaria, neonatal outcome

Introduction

Malaria in pregnancy is a major public health problem with substantial risk to the mother and newborn baby [1]. It imparts negatively on the socio-economic aspect of the environment worldwide. It is a leading cause of death among the susceptible groups in Sub Sahara Africa, 20% of stillbirths, 11% of newborn deaths, and 3.3% of LBW deliveries [2]. Nigeria is not excluded with malaria occurring all year round, contributing to a quarter of malaria in Africa and 300, 000 childhood death [3]. Malaria is associated with an increased risk of miscarriage, maternal anaemia, preterm delivery, stillbirth, low birth weight, intrauterine growth restriction maternal and neonatal mortality fetal anaemia, and congenital malaria [1]. This condition is worsened when coexisting with Human Immunodeficiency Virus (HIV). Globally, the dual infection causes about 4million deaths annually and contributes a significant burden to Sub-Sahara Africa [4]. Nigeria accounts for the second-largest contributor of HIV in Africa and the third-largest in the world [5].

It has been established that the combination alters the natural presentation, pattern and worsened the outcome of either disease when combined [3,4]. Studies has shown that malaria doubles the risk of having HIV, a similar study has attributed 30-50 % of malaria has been linked to HIV infection [5]. Malaria and HIV co-infection have disproportionate effects on pregnant women and pose serious health consequences. In sub-Saharan Africa, a recent estimate has shown that an additional 3 million cases of malaria and 65 000 additional malaria-related deaths annually are due to the impact of HIV [3]. Rates were highest in countries with high HIV prevalence and unstable malaria transmission. Pregnant women with co-infection have more febrile illnesses, worse anaemia and more adverse birth outcomes such as LBW, prematurity and intrauterine growth retardation than women with single infections with either malaria or HIV [6,7]. In Kenya, dual infection was shown to result in a 3-fold increased risk of LBW in primigravidae, and more than doubled the risk of severe to moderate anaemia in both primigravid and multigravid women [8]. Dual infection also increases the risk of maternal, perinatal and early infant death [4].

Maternal malaria parasites in the peripheral blood are sequestered in the placenta and subsequently transferred to the fetus before delivery. This leads to congenital malaria. Congenital malaria is the presence of asexual *P. falciparum* parasites in the cord blood or peripheral blood of an infant within the first 7 days of life. There is a need to determine the presence of malaria on the fetus in determining the neonatal outcome [1,5,9].

The aim of this study was to determine the prevalence and the impact of placental malaria parasitemia on congenital malaria and the birth weight of babies of HIV positive pregnant women in Lagos, Nigeria.

Methodology

Study design: This was a comparative cross-sectional study in which 90 known HIV positive, pregnant women, on HAART, receiving care at the obstetrics and gynaecology department of the Lagos State Teaching Hospital were consecutively recruited at term for assessment of maternal, placenta and congenital malaria at delivery.

Study criteria: Inclusion criteria were consenting, low-risk HIV positive, booked pregnant women with term singleton pregnancy and uncomplicated delivery. Excluded from this study were women with fever or recently treated for malaria in the prior two weeks, women with medical conditions like renal disease, cardiac disease, liver disease hypertension diabetes and haemoglobinopathy or bleeding disorder. Women with history of blood transfusion in the last four weeks of pregnancy and women with AIDS complex or CD4 less than 200 cell/ml were also excluded

Data collection: Every consecutive parturient who satisfied the eligibility criteria, was informed about the study and those who consented were recruited to take part in the study. Data were obtained with the aid of a structured proforma designed for the study by the researchers. Relevant information obtained included age, level of education, religion, parity, booking status, estimated gestational age, HIV status, number of use of Intermittent Preventive Therapy for malaria in pregnancy, and birth weight. Blood samples were collected from the mothers and newborns for packed cell volume (PCV) and malaria parasite testing within two hours of birth. Biopsy and blood sample of the placenta was also obtained to test for placenta malaria and PCV respectively, immediately after delivery. The results of all tests done were noted on the respective proforma assigned to the woman.

Sample collection and analysis: The maternal and neonatal samples were collected by venepuncture using a standard sterile procedure within two hours of delivery. The placental blood was also collected immediately after delivery from the maternal surface of the placenta. An incision was made on the maternal surface of the placenta and placental blood aspirated from the sinuses into an EDTA bottle.

The RDT for malaria was done using the Histidine Rich Protein-2 diagnostic test kit made by Premier Medical Corporation Limited with the trade name First Response. The test kit is based on the principle of immuno-chromatographic in which nitrocellulose membrane is pre-coated with a monoclonal antibody specific to Histidine-Rich Protein 2 (HRP2) produced by Plasmodium falciparum.

The placental tissues were received at the Lagos University Teaching Hospital by a laboratory technician who ensured the samples are properly registered. The placental tissues were processed whole inside a well labeled tissue embedding cassette. Processing was done using a 24 hours automatic tissue processor for an average of 17-19 hours. The automatic tissue processor contains 12 beakers, 10 glass beakers and 2 thermostatically controlled electric metal beakers containing paraffin wax. The first beaker contains 10% formalin for complete fixation, beakers 2-8 contain ascending grades of alcohol from 70% to absolute alcohol so as to remove water from the tissues. Beakers 9 and 10 contain xylene, which clears the alcohol while beakers 11 and 12 contain molten paraffin wax which was used as an embedding agent. Once the specimens were embedded, cut sections were taken using a microtome. The cut sections were placed on clean, grease- free slides which were heated on a hot plate for 30 minutes in order for sections to adhere to the slides. The slides were then stained using Haematoxylin and Eosin (H & E) and Giemsa.

Analysis of placental histology for malaria: The slides were read at Lagos University Teaching Hospital histopathology laboratory under the supervision of a consultant pathologist. A new two-parameter semi- quantitative grading scheme that scores the degrees of inflammation and pigment deposition during placental malaria was adopted, because the new scheme is simple to perform [10]. The application of existing placental malaria grading schemes are limited, because chronic placental malaria is a broad category encompassing varying degrees of pathology, scoring criteria vary by study site, and the degree of inflammation is not independently considered.

The inflammation score is qualitative and is intended to reflect distinct biologic entities, see figure 3A below [10]. A score of (I) is given when there is minimal inflammation which describes cases with no appreciable intervillous inflammation. Pigmented monocytes are rare, and the intervillous space white cell density is not increased above the level of peripheral white blood cells expected in transit. An Inflammatory score of (II) is given when there is moderate inflammation and it describes cases with mononuclear cells sequestering in the intervillous spaces, particularly pigment-laden macrophages. A score of (III) is given when there is massive intervillositis (III) in which the intervillous spaces contain sheets of densely packed mononuclear cells.

The pigment-deposition scoring system is a semi-quantitative method that can be rapidly assessed by quantifying the percentage of high-power fields that are positive for the pigments in fibrin in intervillous spaces (Figure 3B) [11]. The scoring method excludes pigment in erythrocytes or monocytes. Malaria pigment is golden brown after Giemsa staining. The total field count excludes stromal tissue in the decidua, basal plate, and stem villi. Although the total amount of fibrin may be variable, the total field count includes fields in intervillous spaces that do not have fibrin. A score of I is given when less than 10% of the fields are positive, a score of II is given when the estimated pigment is between 10%-40%, while a score of III is given when the pigment is estimated to be over 40% [11,12].

For the purpose of this study, all slides positive for inflammation and pigments are considered positive for placental malaria irrespective of the score.

The data obtained were analyzed using the Statistical Package for Social Sciences, (SPSS) version 19. Percentages, means and standard deviation of numerical variables were determined. Percentages of categorical data were also determined. Means of numerical variables were compared using the student's t-test, while the Chi-square test was used to test for association between categorical variables. The correlation was tested using spearman's correlation. P-value level less than 0.05 was considered statistically significant.

This study was approved by the health research and ethics committee of the Lagos State University Teaching Hospital, Lagos, Nigeria.

Results

Majority (60%) of the HIV positive women who participated in the study were between 25 and 34 years of age (**Table 1**). About 1 in 3 (64.4%) of the women had tertiary education and similar proportions (65.6%) were Christians (**Table 1**). Only 13.3% of the study participants were primigravida (**Table 1**).

The prevalence of malaria at delivery in HIV positive women who were studied was 3.3% in maternal venous blood and 8.9% in maternal placenta blood using the rapid diagnostic test kits for malaria (**Table 2**). The prevalence of placenta malaria in the study was 11.1% (**Table 2**). There was no record of positive malaria blood test at birth in babies born to the women who participated in the study (**Table 2**).

The mean packed cell volume of the neonates at birth of women with placenta malaria was 43.6% and 44.9% in neonates born to women without placenta malaria (p=0.044) (**Table 3**). The mean birth weight of the babies born to HIV positive women was 2.7±0.2kg in those with placenta malaria and 3.0±0.4kg in those without placenta malaria (p=0.008) (**Table 3**).

The correlation between birth weight and CD4 count in these women was positive but insignificant (r=0.069, p=0.515) (**Figure 1**).

Table 1: Characteristics of the Study Participants

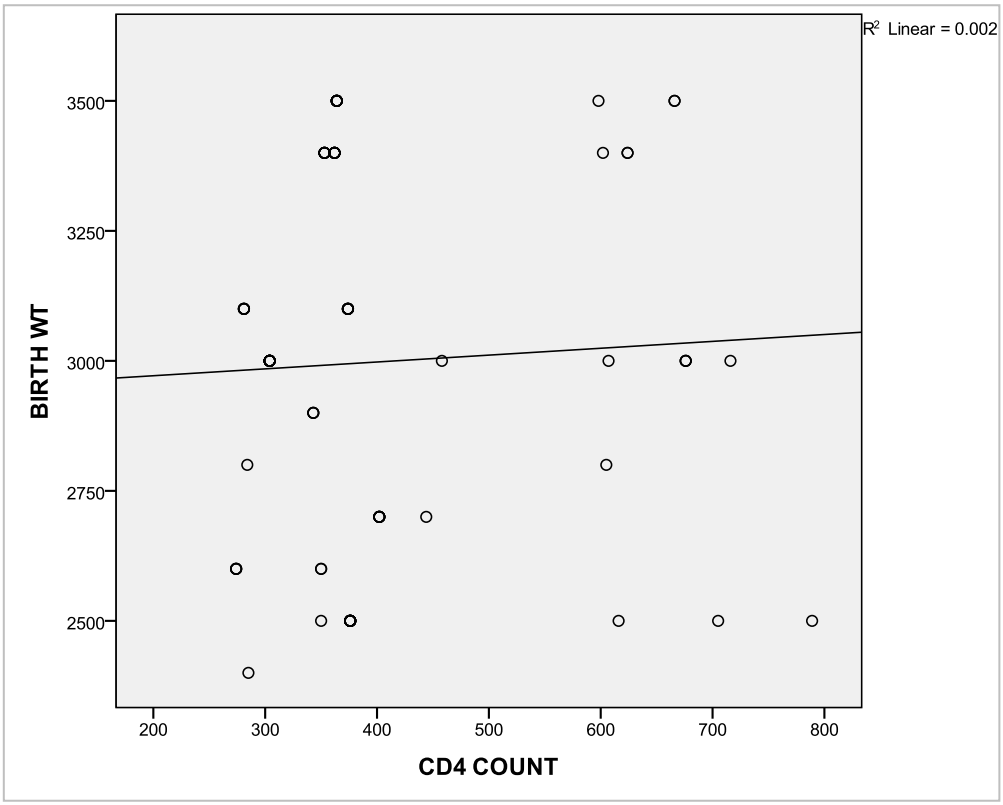
Variable	Frequency (N=90)	Proportion (%)
Age group (years)		
<25	12	13.3
25-34	54	60
≥35	24	26.7
Mean ± SD	30.5 ± 5.0	
Level of Education		
Below Tertiary	58	64.4
Tertiary	32	35.6
Religion		
Christianity	59	65.6
Islam	31	34.4
Gravidity		
Primigravida	12	13.3
Multigravida	78	86.7
SD: Standard deviation		

Table 2: Prevalence of Maternal Placenta and Congenital Malaria

Variable	Frequency (N=90)	Prevalence
Maternal Blood (RDT)		
Positive	3	3.3
Negative	87	
Placenta Blood (RDT)		
Positive	8	8.9
Negative	82	
Placenta Histology		
Positive	10	11.1
Negative	80	
New-born Blood (RDT)		
Positive	0	0.0
Negative	90	
RDT – Rapid diagnostic test for malaria		

Table 3: Comparison of Maternal and New-born Characteristics in HIV positive women based on Placenta Malaria Status

	Placenta Malaria on Histology		
Variable	**Positive n=10 (%)**	**Negative n=80 (%)**	**p-value**
Age group (years)			
< 35	5 (50.0)	61 (76.2)	0.741*
≥ 35 years	5 (50.0)	19 (23.8)	
Parity			
Nulliparous	3 (30.0)	28 (32.5)	1.000*
Multiparous	7 (70.0)	54 (67.5)	
Doses of IPT in Pregnancy			
≤ 2 doses	8 (88.9)	31 (38.6)	0.009*
≥ 3 doses	1 (11.0)	49 (61.3)	
PCV at delivery	30.4±0.7	30.4±0.9	0.933#
Neonatal PCV (%)	43.6±1.3	44.9±2.0	**0.044#**
Birth weight (kg)	2.7±0.2	3.0±0.4	**0.008#**
IPT – Intermittent Preventive Therapy for Malaria administered during pregnancy, PCV- Packed cell volume, kg – kilogram, n – frequency, * - Chi square applied, # - independent t-test applied.			



Spearman Rho correlation coefficient = 0.069, p = 0.515

Figure 1: Correlation between CD4 count and birth weight in HIV positive women

Discussion

The prevalence rate of placental malaria using histological diagnosis in HIV positive parturients in this study was 11.1%. Only 3.3% of the women in our study had a positive peripheral blood malaria test while placenta blood malaria positivity was 8.9%.

This shows that the placenta histology is the most sensitive in the diagnosis of placenta malaria, this has been established has the gold standard for malaria diagnosis. Placenta blood which is the second most sensitive tool from our study reveals the pooling ability of the placenta retain the parasite long after the parasite has disappeared from the peripheral blood [7,11]. Placenta malaria connotes that the malaria infection is either active, chronic or active on chronic [7,13]. The disadvantage of placenta malaria diagnosis is that diagnosis is retrospective, that is after birth. The parasites infected red blood cells using its antigen VARCSA to bind to placenta receptor chrondroitin sulphate A on the fibrin in the placenta enhancing the sequestration in the placenta bed [7,9,11].

Placenta blood using RDT is an easier test, requires no expertise, less cumbersome means of diagnosis, but this requires a larger density of parasitaemia in diagnosis thus in asymptomatic participants in our study would yield a lower positive rate [13]. The drawback is that it requires high malaria density in the blood for a positive result [11].

The peripheral blood using RDT shows that 3.3% of the women were positive. The majority of the parasites is sequestered in the placenta and thus reducing peripheral blood parasitaemia. All the participants were asymptomatic and ensured to be afebrile in the last 2 weeks prior to the study and thus may therefore have low peripheral parasite density. The absence of malaria peripheral blood, although does not connote a negative diagnosis as may be due to the low parasites densities [9,14]. Based on sensitivity RDT is not ideal for making diagnosis further research is therefore needed in making an ideal diagnosis [14,15].

The prevalence of congenital malaria was zero among the babies born to the HIV positive mothers in a malaria-endemic region like ours. Most congenital malaria has been found commonly among babies born to mother travelled from areas of low endemicity to areas of high endemicity [7,16] or primigravida who are yet to generate antibodies to VASRC and thus lack of the maternal antibodies which attack the parasites in the blood [7,9,10,11].

All the babies were negative for malaria parasite when their peripheral blood was tested with RDT in the study, this correlates well with the low result seen by the peripheral RDT result which was 3.3%. However, buttresses the fact that placenta blood and histological are as a result of the pooling effect of the placenta and gives a better sensitivity in determining the diagnosis.

The number of doses of IPT that was significantly associated with placenta malaria status, most women who had more 2 doses were negative for placenta malaria except for one. Babalola et al in Southwest Nigeria found that the risk of malaria was three times higher in women who did not use IPT [11]. Similarly, studies in Uganda and Zambia had found that pregnant women who received one or two doses experienced a five or tenfold reduction, respectively, in the risk of malaria infection [17,18].

The neonatal PCV and birth weight were also significantly associated with placental malaria. Neonates whose mothers had a negative placental malaria result have a higher PCV (44.9±2.0%) and a higher birth weight (3.0±0.4kg) when compared with neonates of mothers, who were positive for placental malaria, 43.6±1.3 % and 2.7±0.2kg respectively. Ayisi et al found similar finding in the neonatal outcomes of women with dual infection [10,19]. Studies have shown that malaria reduces the birth with weight of neonates, this occurs when a high concentration of malaria parasites is sequestered in the placenta [6,7,11]. The sequestration causes several inflammatory changes and infarction in the placenta bed and reduces the blood supply to the fetus [7,11,19,20]. These findings are consistent in babies with congenital malaria [20]. In our study all the babies tested negative for malaria, Eki-udoko et al found significantly lower birth weight in HIV exposed compared to HIV unexposed infants despite the presence of congenital malaria in both groups. They suggested that this may be attributable to maternal HIV status and not necessarily the associated placenta malaria [7]. This differ from our study of significantly lower birth weight when HIV co-existed with

placenta malaria compare to HIV without placenta malaria. There is therefore an obvious need for further research on this topic.

Conclusion

Congenital malaria is a rarity in babies born to HIV positive pregnant women with or without placenta malaria. However, the birth weight and packed cell volume of babies born to HIV positive women with placenta malaria were significantly lower than in HIV positive women without placenta malaria.

Ethics approval and consent to participate

This study was approved by the health research and ethics committee of the Lagos State University Teaching Hospital, Ikeja. The participants were counselled on the research protocol in a language they understood. The participants, having been informed gave an informed consent. The study lasted over a period of seven months

List of abbreviations

HIV: Human immunodeficiency virus
IPT: Intermittent Preventive Therapy for Malaria administered during pregnancy,
PCV: Packed cell volume
RDT: Rapid diagnostic test for malaria
CD4 Count: Cluster of differentials for T cells measurement

Data Availability

The data are readily available

Conflicts of Interest

There is no conflict of interest among authors

Funding Statement

There are no financial support from any source

Authors' Contribution

T. K. Conceived and designed the study, O.O. collected data T. K. wrote the first draft. A. I., F. A. and O. O. participated in the design of the study, interpretation of data. O. O. collected and analysed samples. O.A., O. O., TK critically reviewed for content and all authors approved the final manuscript.

Acknowledgments

Nil

References

[1] Nekaka R, Nteziyaremye J, Oboth P, Iramiot JS, Wandabwa J (2020) Malaria preventive practices and delivery outcomes: A cross-sectional study of parturient women in a tertiary hospital in Eastern Uganda. PLoS ONE 15(8): e0237407

[2] World Health Organization. Implementing malaria in pregnancy programs in the context of World Health Organization recommendations on antenatal care for a positive pregnancy experience. https://www.who.int/reproductivehealth/publications/implementing-malaria-pregnancyprogrammes-brief/en/. Accessed 23 Sep 2019.

[3] Onankpa BO, Jiya NM, Yusuf T. Malaria parasitemiain HIV-infected children attending antiretroviral therapy clinic in a teachinghospital. Sahel Med J 2017;20:30-2.

[4] Nteziyaremye J, Oboth P, Iramiot JS, Wandabwa J (2020) Malaria preventive practices and delivery outcomes: A cross-sectional study of parturient women in a tertiary hospital in Eastern Uganda. PLoS ONE 15(8): e0237407

[5] Awofala AA, Ogundele OE. HIV epidemiology in Nigeria. Saudi J Biol Sci. 2018;25(4):697-703. doi:10.1016/j.sjbs.2016.03.006

[6] Park, S., Nixon, C. E Miller, O., Choi, N. K., Kurtis, J. D., Friedman, J. F., & Michelow, I. C. (2020). Impact of malaria in pregnancy on risk of malaria in young children: Systematicreview and meta-analyses. Journal of Infectious Diseases

[7] Edo State Eki-udoko E Fidelis et al Placental Malaria histological features and the burden of congenital malaria among HIV/Malaria Co-infected Mothers in Benin City, Niger J Paediatr 2021; 48 (1): 12 - 19

[8] Izuka EO, Ugwu EO, Obi SN, Ozumba BC, Nwagha TU, Obiora-Izuka CE. Prevalence and predictors of placental malaria in human immunodeficiency virus-positive women in Nigeria. Niger J Clin Pract 2017;20:31-6

[9] Solomon, A., Kahase, D. & Alemayhu, M. Prevalence of placental malaria among asymptomatic pregnant women in Wolkite health center, Gurage zone, Southern Ethiopia. Trop Dis Travel Med Vaccines 6, 20 (2020).

[10] Ayisi JG, van Eijk AM, ter Kuile FO, Kolczakc MS, Otieno JA, Misore AO, Kager PA, Steketee RW, Nahlen BL. The effect of dual infection with HIV and malaria on pregnancy outcome in western Kenya, AIDS, 2003; 17: 585–594

[11] Babalola et al. Malaria infection at parturition in Abeokuta, Nigeria: Current status and pregnancy outcome MWJ 2017, 8:12

[12] Muehlenbachs A, Fried M, McGready R, Harrington WE, Mutabingwa TK, Nosten F, et al. A novel histological grading scheme for placental malaria applied in areas of high and low malaria transmission. The Journal of infectious diseases 2010;202(10):1608-1616.

[13] Liu Y, Griffin JB, Muehlenbachs A, et al. Diagnosis of placental malaria in poorly fixed and processed placental tissue. Malar J. 2016;15:272

[14] Kyabayinze DJ, Zongo I, Cunningham J, et al. HRP2 and pLDH-Based Rapid Diagnostic Tests, Expert Microscopy, and PCR for Detection of Malaria Infection during Pregnancy and at Delivery in Areas of Varied Transmission: A Prospective Cohort Study in Burkina Faso and Uganda. PLoS One. 2016;11:e0156954 .

[15] Liu Y, Mwapasa V, Khairallah C, et al. Rapid Diagnostic Test Performance Assessed Using Latent Class Analysis for the Diagnosis of Plasmodium falciparum Placental Malaria. Am J Trop Med Hyg. 2016;95:835-839

[16] Melissa Bauserman MD MPH, Andrea L. Conroy PhD, Krysten North MD MPH, Jackie Patterson MD MPH, Carl Bose MD, Steve Meshnick MD PhD, An Overview of Malaria in Pregnancy, Seminars in Perinatology (2019), doi: https://doi.org/10.1053/j.semperi.2019.03.018.

[17] De Beaudrap, P., Turyakira, E., White, L.J., Nabasumba, C., Tumwebaze, B., Muehlenbachs, A., Guérin, P.J., Boum II, Y., McGreadyand, R. and Piola, P. Impact of Malaria during Pregnancy on Pregnancy Outcomes in a Ugandan Prospective Cohort with Intensive Malaria Screening and Prompt Treatment. Malaria Journal,2013: 12, 139.

[18] Mace, K.E., Chalwe, V., Katalenich, B.L., Nambozi, M., Mubikayi, L., Mulele, C.K., Wiegand, R.E., Filler, S.J., Kamuliwo, M., Craig, A.S. Tan, K.R. Evaluation of Sulphadoxine-Pyrimethamine for Intermittent Preventive Treatment of Malaria in Pregnancy: A Retrospective Birth Outcomes Study in Mansa, Zambia. Malaria Journal, 2015:14, 69

[19] Otuli Noël L, Nguma Jean-Didier B, Alongo Mike-Antoine M, Bosunga Gedeon K, Mukonkole Jean-Paulin M, Likwela Joris L, Okenge Jean-Pascal M. Prevalence of Congenital Malaria in Kisangani, A Stable Malaria Transmission Area in Democratic Republic of the Congo. Infect Dis Obstet Gynecol. 2020, 25;2020:2176140. doi: 10.1155/2020/2176140. PMID: 32158176; PMCID: PMC7060851.

[20] L.L Chua, W Hansang, S.J. Rogerson, A Teo. Poor Birth Outcomes in Malaria in Pregnancy: Recent Insights into Mechanisms and Prevention Approaches cl Front. Immunol., 2021 :15 |