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THE EFFECT OF MALARIA INFECTION ON SOME HEMATOLOGICAL PARAMETERS OF PREGNANT WOMEN IN NNEWI, ANAMBRA STATE, NIGERIA

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ABSTRACT

This study was designed to assess the effect of malaria infection on some haematological parameters of pregnant women in Nnewi, Anambra State, Nigeria. A total of 100 participants aged between 18 and 36 (25±4) years were randomly recruited for the study. The participants were grouped as follows: 50 pregnant women and 50 non pregnant women. Thereafter, 5mls each of blood samples were collected from the participants for the determination of malaria parasites, hemoglobin (Hb), packed cell volume (PCV), and white blood cell (WBC) levels using standard laboratory methods. The results showed that Hb and PCV levels were significantly lower in pregnant women and malaria infected pregnant women respectively ($p<0.05$). However, neutrophil levels were significantly higher in pregnant women as well as malaria infected pregnant women than in the control respectively ($p<0.05$). Also, WBC and neutrophil levels were significantly higher in malaria infected pregnant women in their first trimester than in control respectively ($p<0.05$). The implication of these findings is that both malaria infection and pregnancy have effect on some haematological parameters in women.

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INTRODUCTION

Malaria is a mosquito-borne infectious disease of humans caused by a protozoan parasite of the genus called plasmodium. It is transmitted through the bite of infected female anopheles mosquitoes (in the human body by blood meal) from its saliva into a person's circulatory system. In the blood, the parasite travels to the liver where it matures and reproduces; multiplying in the liver and then infects the red blood cells and causes symptom that typically include fever and headache, in severe cases progressing to coma and death (Robert *et al*., 2002). Malaria can be caused majorly by *P. falciparium,P. malariae*, *P. ovale* and *P. vivax* (Collins, 2012). Malaria during pregnancy is a major public health problem; it increases the risk of low birth weight (<2500g), still birth, infant mortality and morbidity during the first year of life by inducing intra-uterine growth retardation, prematurity and infant anemia (Hartman *et al*., 2010). Malaria is the most prevalent infectious disease in the tropical and sub-tropical regions of the world in addition to being the major cause of morbidity in the tropics (Mishral *et al.,* 2003; Mia *et al*., 2011). This is because of the significant amounts of rainfall and consistent high temperatures and high turbidity along with stagnant waters which provide mosquitoes the favourable environment needed for continuous breeding (Prothero and Mansell, 1999). The World Health Organisation reported that malaria is responsible for nearly 90% of deaths in Africa (Ogbodo *et al*., 2010) while records have shown that about 50% of the Nigerian population suffers from at least one episode of malaria annually with over 45% of all out-patient visits being associated with malaria (Ejezie *et al*., 1991; Federal ministry of Health, 2001). The prevalence of malaria is

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higher during pregnancy compared with the non-pregnant state (Giles *et al.,* 2000). Malaria infection in pregnant women is associated with high risk of both maternal and prenatal morbidity and mortality. Pregnant women have reduced immune response and therefore unable to effectively clear malaria infections (Elliot *et al.,* 2005). Malaria infection is more frequent in primigravidae and secundigravidae than in multigravidae (Van Geertruyden, 2005). Malaria is typically diagnosed by the microscopic examination of blood using blood films or with antigen-based rapid diagnostic tests (Kattenberg *et al*., 2011). In areas of stable malaria transmission where the most common species is *P. falciparum,* most infections have symptoms like shivering, joint pain, hemolytic anemia (Guyatt and Snow, 2001), hemoglobin in the urine, retinal damage, convulsion, jaundice, vomiting, fever, headache and usually persist for long periods at low densities (Beare *et al*., 2006). Furthermore, malaria parasites sequester and replicate in the placenta (Luxemburger et al., 2005), the malaria infection during pregnancy can lead to miscarriage, premature delivery, low birth weight, congenital malaria infection and perinatal death amongst others (Mutabingwa *et al*., 2005). Also, in Nigeria, malaria causes a lot of debilitating effects in adults but has been neglected (Major *et al.,* 2007) and yearly, economic loss due to malaria amounts to 132 billion naira (Federal Ministry of Health, 2005). Again, there is paucity of information on the changes in the hematological parameters of symptomatic adults (pregnant and non-pregnant women) with malaria infection without any other disease conditions. Therefore, this study is designed to evaluate the effect of malaria infection on some hematological parameters of pregnant women in Nnewi, Anambra State, Nigeria.

MATERIALS AND METHODS

Research design and sample collection

This is an experimental study designed to assess the effect of malaria infection on some haematological parameters on pregnant women in Nnewi, Anambra State, Nigeria. A total of 100 participants aged between 18 and 36 years were randomly recruited for the study. The participants were grouped into two comprising of 50 pregnant women and 50 non pregnant women. Thereafter, 5mls of blood sample each was collected from the subjects into EDTA bottles for the determination of malaria parasites, hemoglobin level, packed cell volume and white blood cell count.

Inclusion and exclusion criteria: Pregnant and non pregnant women aged between 18 and 36 years were included for the study while the study excluded children, women younger than 18 or above 36 years, women who were sick and those already on anti-malaria drug.

Determination of haematological parameters: White blood cell count (WBC), packed cell volume (PCV) and hemoglobin level was determined using Mythic 22 automated hematology analyzer.

Diagnosis of Malaria: Thick and thin films were prepared and stained with Giemsa stain for parasite identification and quantification using standard methods as described by WHO (1995), assuming a leukocyte count of $8000\mu L^{-1}$. Films were examined microscopically for the presence of malaria parasites within red blood cells in thin films whereas, the ring forms, trophozoites and gametocytes were noted for in thick films. A smear was considered negative for malaria parasites if no parasites are seen after examining at least 100 microscopic fields.

Ethical Consideration: This was sought and obtained from Faculty of Health Sciences and Technology Ethical Committee, Nnewi. Informed consent was obtained from participants prior the commencement of the study.

Statistical analysis: Data obtained were tabulated and analyzed using SPSS version 20.0 (SPSS Inc. Chicago, IL, USA). Student's t-test was employed in comparing means and results expressed as Mean±SD. $P<0.05$ was considered statistically significant.

RESULTS

The mean blood haemoglobin level (g/dl), PCV (L/L) and age were significantly lower in pregnant women compared to the control (p=0.000), but no statistical significant difference was observed in white blood cell count (mm^3) compared between both groups (p=0.646) (See Table 1). In this study, the mean level of neutrophil was significantly higher in pregnant women compared to the control (p=0.000). However, the mean levels of lymphocytes, monocytes and eosinophils did not differ significantly (p>0.05) (See Table 2). Between group comparisons, the mean blood haemoglobin and packed cell volume were significantly lower in malaria infected pregnant

Table 1. Comparison of mean (±SD) WBC, Hb, and PCV levels in pregnant and non pregnant women in Nnewi

Variables	WBC (mm^3)	Hb (g/dl)	PCV (%)	Age (yrs)
Pregnant women (n=50)	6198.20±2111.40	8.87±1.70	34.32±5.59	27.32±4.42
Control (n=50)	6029.20±1500.65	10.53±1.62	41.48±4.93	22.72±4.67
t-value	3.121	0.489	2.011	2.639
p-value	0.646	0.000*	0.000*	0.000*

*Statistically significant at p<0.05.

Table 2. Comparison of mean (±SD) lymphocyte, monocyte, eosinophil and neutrophil levels in pregnant and non pregnant women in Nnewi

Variables	Lymphocyte (µl)	Monocyte (µl)	Eosinophil (µl)	Neutrophil (µl)
Pregnant women (n=50)	30.14±4.15	5.42±1.69	2.72±1.05	62.24±3.62
Control (n=50)	32.14±6.59	5.94±1.68	2.58±1.07	57.08±4.68
t-value	14.720	0.382	0.150	0.719
p-value	0.127	0.127	0.453	0.000*

*Statistically significant at p<0.05.

Table 3. Comparison of mean (±SD) WBC, Hb, and PCV levels in malaria infected pregnant women and malaria negative control

Variables	WBC (mm^3)	Hb (g/dl)	PCV (%)	Age (yrs)
MP (+) Pregnant women (n=40)	6535.20±2165.71	8.86±1.77	34.18±5.35	27.00±4.52
MP (-) Control (n=15)	5840.70±1847.86	10.31±1.70	41.40±4.49	22.13±4.63
t-value	0.027	0.213	1.437	0.013
p-value	0.027	0.003*	0.000*	0.000*

*Statistically significant at p<0.05.

Table 4. Comparison of mean (±SD) lymphocyte, monocyte, eosinophil and neutrophil levels in malaria infected pregnant women and malaria negative control

Variables	Lymphocyte (µl)	Monocyte (µl)	Eosinophil (µl)	Neutrophil (µl)
MP (+) Pregnant women (n=40)	30.03±4.44	5.30±1.71	2.68±1.02	62.75±3.66
MP (-) Control (n=15)	29.33±6.84	6.33±1.35	2.73±1.16	58.07±3.49
t-value	8.289	2.492	1.379	2.812
p-value	0.661	0.040*	0.857	0.002*

*Statistically significant at p<0.05.

Table 5. Comparison of mean (±SD) WBC, Hb, and PCV levels in malaria infected pregnant women and malaria negative control in first trimester

Variables	WBC (mm^3)	Hb (g/dl)	PCV (%)	Age (yrs)
MP (+) Pregnant women in first trimester (n=25)	7116.40±2479.19	8.99±1.60	34.40±4.55	28.56±4.37
MP (-) Control (n=8)	4806.20±1219.61	9.63±1.04	35.63±7.37	26.63±4.44
t-value	4.865	2.478	1.922	1.588
p-value	0.017*	0.301	0.575	0.286

*Statistically significant at p<0.05.

Table 6. Comparison of mean (±SD) lymphocyte, monocyte, eosinophil and neutrophil levels in malaria infected pregnant women and malaria negative control in first trimester

Variables	Lymphocyte (µl)	Monocyte (µl)	Eosinophil (µl)	Neutrophil (µl)
MP (+) Pregnant women in first trimester (n=25)	30.16±5.06	5.56±1.73	2.72±0.94	62.72±3.62
MP (-) Control (n=8)	30.50±3.12	6.13±1.64	3.13±1.25	59.75±2.71
t-value	2.930	0.082	0.206	2.166
p-value	0.860	0.423	0.333	0.042*

*Statistically significant at p<0.05.

Table 7. Comparison of mean (±SD) WBC, Hb, and PCV levels in first trimester and second trimester

Variables	WBC (mm^3)	Hb (g/dl)	PCV (%)	Age (yrs)
first trimester (n=33)	6556.40±2438.43	9.14±1.49	34.70±5.28	28.09±4.40
Second trimester (n=11)	5886.40±917.09	7.89±1.47	32.09±6.24	25.64±4.43
t-value	7.019	0.090	0.242	1.035
p-value	0.381	0.020*	0.181	0.117

*Statistically significant at p<0.05.

women than in the control (p=0.003; 0.000) respectively. Also, the level of white blood cell was significantly higher in malaria infected pregnant women than in control (p=0.027) (See Table 3). The result showed that the mean level of neutrophil was significantly higher in pregnant women with malaria parasitemia compared to the control group (p=0.002), whereas the mean level of monocyte was significantly lower in pregnant women with malaria infection than in control group (p=0.040). However, no significant differences were observed in the mean levels of lymphocyte and eosinophil respectively (p>0.05) (See Table 4). Furthermore, the mean level of white blood cell was significantly higher in pregnant women with malaria infection in their first trimester compared to non infected pregnant women in their first trimester (p=0.017), but their mean haemoglobin and PCV levels did not differ significantly (p>0.05) (See Table 5). Also, the present study showed that the neutrophil level was significantly higher in pregnant women with malaria infection in their first trimester compared to non infected pregnant women in their first trimester (p=0.042), while no statistical significant differences were observed in the mean levels of lymphocyte, monocyte and eosinophil respectively (p>0.05) (See Table 6).

Again, the result showed that the blood haemoglobin level was significantly higher in the first trimester compared to those in second trimester (p=0.020), while the mean levels of white blood cells and packed cell volume were similar between both groups (p>0.05) (See Table 7). Similarly, between the group comparison showed no significant difference in levels of lymphocyte, monocyte, eosinophil and neutrophil between first and second trimester respectively (p>0.05) (See Table 8). In this study, there were no significant differences in the mean levels of white blood cells, haemoglobin and packed cell volume between the first and third trimesters respectively (p>0.05) (See Table 9).Also, there were no significant differences in the mean levels of lymphocyte, monocyte, eosinophil and neutrophil in first trimester compared to the third trimester respectively (p>0.05) (See Table 10). More so, there were no significant differences in the mean levels of haemoglobin and packed cell volume in second and third trimester (p>0.05) (See Table 11). Also, there were no significant differences in the mean levels of lymphocyte, monocyte, eosinophil and neutrophil in second trimester compared to the third trimester respectively (p>0.05) (See Table 12).

Table 8. Comparison of mean (±SD) lymphocyte, monocyte, eosinophil and neutrophil levels between first trimester and second trimester

Variables	Lymphocyte (µl)	Monocyte (µl)	Eosinophil (µl)	Neutrophil (µl)
first trimester (n=33)	30.24±4.62	5.70±1.70	2.82±1.01	62.00±3.62
Second trimester (n=11)	29.09±3.27	4.82±1.66	2.55±1.29	63.45±3.91
t-value	0.851	0.334	1.086	0.269
p-value	0.450	0.144	0.475	0.264

*Statistically significant at p<0.05.

Table 9. Comparison of mean (±SD) WBC, Hb, and PCV levels in first trimester and third trimester

Variables	WBC (mm^3)	Hb (g/dl)	PCV (%)	Age (yrs)
first trimester (n=33)	6556.40±2438.43	9.14±1.49	34.70±5.28	28.09±4.40
Third trimester (n=6)	4800.00±738.92	9.22±2.63	36.33±5.89	26.17±4.07
t-value	4.905	0.090	0.242	1.035
p-value	0.091	0.922	0.495	0.326

*Statistically significant at p<0.05.

Table 10. Comparison of mean (±SD) lymphocyte, monocyte, eosinophil and neutrophil levels between first trimester and third trimester

Variables	Lymphocyte (µl)	Monocyte (µl)	Eosinophil (µl)	Neutrophil (µl)
first trimester (n=33)	30.24±4.62	5.70±1.70	2.82±1.01	62.00±3.62
Third trimester (n=6)	31.50±2.35	5.00±1.55	2.50±0.84	61.33±3.01
t-value	1.750	0.135	1.086	0.269
p-value	0.522	0.357	0.474	0.674

*Statistically significant at p<0.05.

Table 11. Comparison of mean (±SD) WBC, Hb, and PCV levels between second trimester and third trimester

Variables	WBC (mm^3)	Hb (g/dl)	PCV (%)	Age (yrs)
Second trimester (n=11)	5886.40±917.09	7.89±1.47	32.09±6.24	25.64±4.43
Third trimester (n=6)	4800.00±738.92	9.22±2.63	36.33±5.89	26.17±4.07
t-value	4.905	5.353	0.000	0.011
p-value	0.091	0.198	0.192	0.812

*Statistically significant at p<0.05.

Table 12. Comparison of mean (±SD) lymphocyte, monocyte, eosinophil and neutrophil levels between second trimester and third trimester

Variables	Lymphocyte (µl)	Monocyte (µl)	Eosinophil (µl)	Neutrophil (µl)
Second trimester (n=11)	29.09±3.27	4.82±1.66	2.55±1.29	63.45±3.91
Third trimester (n=6)	31.50±2.35	5.00±1.55	2.50±0.84	61.33±3.01
t-value	0.723	0.011	1.697	1.100
p-value	0.134	0.829	0.940	0.268

*Statistically significant at p<0.05.

DISCUSSION

Malaria is an important cause of death and illness in tropical countries such as Nigeria and is of paramount public health importance (Trampuz *et al*., 2003). The present study revealed a significantly lower level of haemoglobin and packed cell volume in pregnant women than control group and the implication may be as a result of pregnancy which may result to anaemia (Desai *et al*., 2007). Also, pregnant women with malaria infection were found to have significantly lower levels of haemoglobin and packed cell volume compared with the non-malaria infected group. This is in line with previous studies (Mbanefo *et al*., 2009; Ogbodo *et al*., 2010; Kayode *et al*., 2011; Kotepui *et al*., 2014; Haruna and Daskum, 2018). This may be due to the resultant effect of malaria infection. *P. falciparium* is one of the cardinal causes of anaemia and is the most prevalent species of plasmodium in the study area. Therefore, this study confirms the fact that anaemia is an intrinsic feature of malaria which is more common during pregnancy due to increased susceptibility of this group to malaria and other infections during pregnancy, owing to the suppression of the immune system in order to ensure the establishment and non rejection of the foetus as a foreign allograft (Akanbi *et al*., 2004).

This low levels of PCV and Hb may cause dizziness and fainting in pregnancy and are common signs and symptoms associated with malaria (Nadjim and Brehens, 2012). In this study, the neutrophil level was significantly higher in both apparently healthy pregnant women and malaria infected pregnant women respectively than in the control group. This is in consonance with previous studies (Bostrom *et al*., 2017). Neutrophil possess prominent immune regulatory activities and are the first responders of inflammatory cells to migrate towards the site of inflammation (Cohen *et al*., 2002). It may also be due to the fact that neutrophil fights against antigen since foetus is a foreign body (Clark *et al*., 1986) and also, its high level may be due to the presence of malaria infection in pregnancy. Further, the monocyte level was significantly lower in malaria infected pregnant women than in control. This agrees with the report of Kotepui *et al*., (2014). However, the mean levels of both lymphocyte and eosinophil were similar between both groups. Expectedly, there was a significant rise in white blood cell levels of the pregnant women infected with malaria than control. This is in concert with the report of Adesina *et al*. who reported that WBC increases with rise in the parasite density in Ilorin, Nigeria (Adesina *et al.,* 2009). The age of pregnant women used for this study is a risk factor for malaria and this may be as a result of immunity level. This

is in accordance with the findings made by Dicko *et al*. that younger maternal age are found to be more susceptible to malaria and they are an independent risk factor for malaria in pregnancy due to continuous development of malaria immunity in older women (Dicko *et al*., 2003). The haemoglobin level was significantly higher in first trimester than in second trimester and this is perhaps because haemologbin carried oxygen (Erich, 1995), and the rate of the oxygen transportation may be lower in first trimester compared to the second trimester.

Conclusion

This study revealed significant alterations in the haematological parameters under consideration. Therefore, the implication of this finding is that both malaria infection and pregnancy have effects on some haematological parameters in women and hence caution is necessary in diagnosis, management and treatment of malaria infected pregnant women.

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