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MALARIA IN PREGNANCY A COMPARATIVE STUDY OF PREGNANT WOMEN IN TWO HOSPITALS

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ABSTRACT

The prevalence of malaria among pregnant women was determined in Braithwaite Memorial Specialist Hospital (BMSH) and Bori General Hospital (BGH) by direct examination of stained blood smear using Giemsa method. A total of 200 pregnant women were examined from each hospital and 100 non-pregnant women were used as control subjects. The prevalence of malaria among pregnant women was 27.5% and 35.0% for BMSH and BGH respectively. The prevalence of malaria among non-pregnant subjects was 15.0%. The prevalence of malaria by educational status showed that those with primary/secondary education had prevalence of 10% and 28% from BMSH and BGH respectvely, while those with tertiary education had prevalence of 17.5% and 6.5% in BMSH and BGH respectively. The prevalence of malaria among pregnant women using insecticide treated nets (INTs) was 21% and those who do not 6.5%, from BMSH, whereas from BGH, non-user of INTs had prevalence of 30.0% and user 5.0% respectively. The prevalence of malaria by ABO blood group among pregnant women in both hospitals showed that O had high level parasitemia, (BMSH 16.5% and BGH 19.0%) respectively. The prevalence of malaria by haemoglobin genotype, AA had high infestation rate compared to genotype AS. Measures aimed at controlling and eliminate malaria should be intensified, especially the use of INTs orLLNs that has been shown to reduce the prevalence of malaria; considering the burden malaria places on pregnant women and fetus in malaria endemic areas.

KEYWORDS: Comparative, Study, Malaria, Pregnancy.

INTRODUCTION

Malaria, a parasitic infection transmitted by a protiozoan of the phylum *arthropoda* in the class insecta, order diptera, family culcididae and genus *Anopheles gambiae* (Mayers *et al.,* 2014). Plasmodium infection which is associated with malaria have five species but *P. falciparum* is the most common malaria parasite in Africa and it is associated with more serious health complications such as cerebral malaria, severe anaemia, respiratory acidosis, acute renal failure etc. (Kolawale *et al.,* 2006). Malaria has caused serious morbidity and mortality in Sub-Sahara Africa, with various negative socio-economic effects (WHO, 2001).

WHO estimates there were 214 million new cases of malaria worldwide in 2015 (range 147-303 million). The African Region accounted for most global cases of malaria (88%), South East Asia Region (10%) and the Eastern Mediterranean Region (2%) respectively. In 2015, there were 438,000 estimated malaria deaths (range 236,000-635,000) worldwide, most of the deaths occurred in African Region (90%), followed by South East Asia Region (7%) and Eastern Mediterranean Region (2%)) respectively (WHO,2015).

There had been resurgence of malaria globally, the factors implicated were, re-infection, incomplete or inadequate treatment due to drug resistance or improper choice of medication, antigenic variation, multiple infection by different strains, resistance of parasite to anti-malarial, changes in environment which encourages new breeding sites for mosquitoes and movement (voluntary or due to unrest or war) (Miller *et al.,* 1994; Sowunmi *et al.,* 1997; Chin, 2000).

Malaria affects more than 3,000,000 pregnant women yearly in the developing countries, and it causes poor birth outcomes, maternal anemia and death. Malaria infection in pregnancy increases RBCs destruction and decrease erythropoesis (Huddle *et al.,* 1999; Menendez *et al.,* 2000). Malaria in pregnancy is believed to account for about 25% of severe maternal anemia, 10 – 20% of neonatal anemia and infant deaths due to low birth weight (Greenwood *et al.,* 2005). Recent world malaria report shows that Nigeria accounts for a quarter of all malaria cases in malaria endemic countries in Sub-Sahara Africa, this shows the challenges malaria poses on Nigeria. This might be due to population (150 million) and 1% of maternal death is attributed to malaria (Imeausen *et al.,* 2005). The Federal Ministry of Health

had documented malaria associated maternal death at 11% and 70.5% of morbidity in Nigerian pregnant women. Yearly, about 25,000,000 women are pregnant in Sub- Africa, a region high endemic for malaria (WHO, 2004). This region accounts for about 80 – 90% of global malaria (Guyatt and Snow, 2001). In economic terms, malaria causes huge economic loss (Gallup and Sachs, 2001).

Malaria is dangerous in pregnant women because of reduced immunity associated with pregnancy, this makes them more susceptible to malaria infection and consequences such as maternal anaemia, spontaneous abortion, pre-term labour, stillbirth, placenta infection and death (Curtis *et al.*, 2003).

The proportion of population sleeping under insecticide treated nets (INTs) increased from less than 2% in 2000 to 55% in 2015. Ensuring access to INT is a challenge to the increasing proportion of population sleeping under ITN. Endemic areas like the Sub-Saharan Africa has received over 500 million ITNs between 2013 to 2015, and the proportion that have access to INTs has increased to 67% in 2015 (WHO, 2015). For indoor residual spraying (IRS), the Sub-Sahara Africa has the largest proportion of population at risk of protection by IRS. The proportion at risk declined globally from 5.7% in 2010 to 3.4% in 2014 (WHO,2015). Increased progress have been made in the control of malaria vector in Sub-Sahara Africa mainly using long lasting insecticidal bed nets (LLINs) and indoor residual spraying (IRS).

In Sub-Sahara Africa, blood group O in ABO blood group is between 47-87%, about 47% in Namibia and Ghana and 52-87% in other Sub-Sahara Africa countries (Christine & Walter, 2007). Severe malaria were observed among patients with blood group O, therefore no association between blood groups and malaria (Ann *et al*., 2009). Blood group O subjects with complicated malaria had low parasitemia compared to other blood groups, blood group O individual have survival advantage over other groups in complicated malaria (Richmond *et al*., 2016). Blood group O individuals have more prevalence of malaria compared to other blood groups but less severe or complicated malaria (Amala & Nwigbani, 2015).

The association between haemoglobin genotype and plasmodium infection, genotype HbAA was significantly infected compared HbAS. HbAA were more susceptible to malaria parasitemia than HbAS because of the defective Hb in sickle cell individuals (Akhigbe *et al*., 2011).

MATERIALS AND METHODS

Study Area: This study covered two Local Government Areas; Port Harcourt and Bori both in Rivers State, Nigeria. Port Harcourt is located on latitude 4049127"N and $7^{0}2^{1}1^{11}$E with an area of 109km^{2,} and population of about 2 million. Bori is located on latitude $4^{0}40^{1}22^{11}$ and $N7^{0}22^{1}13^{11}$ with an area of 560km^{2} and population of about 10,000 persons. Both towns have tropical wet climate with lengthy rainy season and short dry season. The average temperature is about 21.3^{0} for both towns.

Study Subjects: A total of 400 pregnant women 200 from each hospital between ages 18-39 years, who were attending antenatal clinics in both hospitals and 100 non pregnant women used as control subjects were recruited for this study. Structured questionnaires were administered to the pregnant women to obtain their demographic data. Pregnant women with fever, weakness, anorexia, those who have taken antimalarial drugs recently and HIV positives were excluded from this study.

The pregnant women were grouped according to their educational status. Those **Collection of samples**: Each subject was positioned and tourniquet was tied at arm to expose the cubital vein. The site for collection of blood sample was sterilized by cleaning with 70% ethanol on cotton wool swab, and using 5ml syringe, blood was collected by venipuncture and transferred into EDTA bottle and mixed properly to prevent coagulation.

Sample Analysis: Samples were analyzed using thick and thin blood films.

Thick Film Preparation: Thick blood films were made by pipetting 12uL of blood onto clean grease-free slide, about 1cm away from the edge of the slide and spread with a slide to make thick smear. The films were air dried.

Thin Film: The thin films were made by pipetting 3uL of blood onto a clean grease free slide, 1cm from the edge. A clean glass slide spreader held at angle 45^{o} was placed on the blood and the blood was allowed to spread along the entire edge of the slide. The slide is push forward rapidly and smoothly to obtain feathered edge.

Staining method: Giemsa staining methods for thick and thin film are carried out according to (Cheesbrough, 2000).

Microscopic examination for malaria parasites: The thick and thin films were examined microscopically using oil immersion (x 100) objective lens.

Determination of ABO blood group: The tile grouping method was used to determine ABO blood group. A drop of test sample (blood) was dropped onto different spots labeled A, B. AB on a white tile. The antisera A, B, AB and D were added to the corresponding spots. They were mixed properly with an applicator stick and examined for agglutination both macroscopically and microscopically. Agglutination reaction signified the presence of natural antigen A, B, AB or D. Determination of Haemoglobin genotype (Hb-genotype) was done by electrophoresis as described by (Chessbrough, 2000).

Statistical analysis: Statistical analysis was done using the statistical package for social science (SPSS).

RESULTS

The prevalence of malaria among pregnant women attending antenatal clinics in BMSH was 27.5%, while the prevalence among those attending BGH was 35.0% respectively. The overall prevalence of malaria among pregnant women from both health facilities was 31.2%. The prevalence of malaria was slightly high among pregnant women attending BGH by 7.5% as shown in table 4.1.

Table. 4.1: Prevalence of malaria among pregnant women attending antenatal clinics in BMSH and BGH.

Hospital	Number of Samples examined	Number positive
BMSH	200	55(25.7)
BGH	200	70(35.0)
CONTROL	100	15(15.0)
Total	**400**	**125(35.2)**

Numbers in parenthesis = percentages

Those, Who had primary and or secondary education in BMSH were 60, 20(10.0%) had malaria, while those with tertiary education were 140, 35(17.5%) had malaria respectively. Whereas in BGH, 149 had only primary or both primary/ secondary education 57(28.5%) had malaria; 51 had tertiary education, 13(6.5%) had malaria. In both hospital, 209 pregnant women had primary/secondary education 77(19.3%) had malaria and those with tertiary education were 191, 48(12.0%) were infected respectively.

Table. 4.2 Prevalence of malaria among pregnant women by educational status.

	BMSH		BGH	
Educational Status	**Number examined**	**Number positive**	**Number examined**	**Number positive**
Primary/secondary	60(30)	20(10.0)	149(74.5)	57(28.3)
Tertiary	140(70)	35(17.5)	51(25.5)	13(6.5)

Numbers in parenthesis=percentages.

The prevalence of malaria among pregnant women using insecticide treated nets (INTs) from BMSH was 54(27%) and 13(6.5%) were infected; 146(73%) non users of INTs, 42(21.0%) were infected with malaria parasites. In BGH, 49 used INTs 10(5.0%) were infected, whereas 151 do not use INTs, 60(30.0) were infected. The overall prevalence among pregnant women that use INTs was 7.5%, whereas non INTs user was 25.5% in both hospitals.

Table. 4.3 Prevalence of malaria among pregnant women using INTs and non- INTs users.

	BMSH		BGH	
Educational Status	**Number examined**	**Number positive**	**Number examined**	**Number positive**
Users of INTs	54(27)	13(6.5)	49(24.5)	10(5.0)
Non INTs users	146(73)	42(21.0)	151(75.5)	60(30.0)

Numbers in parenthesis=percentages.

Prevalence of malaria among pregnant women by ABO-blood groups showed that, blood group O had high infestation compared to other ABO-blood groups. In BMSH, blood group O were 104(52%), 33(16.5%) were infected, blood group A were 60(30%), 15(7.5%) were infected, B 32(16%), 5(2.3%) were infected and AB 4(2%), 2(1%) infected respectively. In BGH, O were 106(53%), 38(19.0%) were infected, A 54(27%), 22(11.0%) were infected, B, 34(17%), 9(4.5%) were infected and AB 6(3%), 1(0.5%) had malaria parasites respectively.

Table. 4.4 Prevalence of malaria among pregnant women by ABO blood group.

	BMSH		BGH	
Blood Group	**Number examined**	**Number positive**	**Number examined**	**Number positive**
O	104(52)	33(16.5)	106(53)	38(19.0)
A	60(30)	15(7.5)	54(27)	22(11.0)
B	32(16)	5(2.5)	34(17)	9(4.5)
AB	4(2)	2(1.0)	6(3)	1(0.5)
Total	**200**	**55(27.3)**	**200**	**70(35.6)**

Numbers in parenthesis=percentages

The prevalence of malaria by hemoglobin genotype among pregnant women showed that about 81 – 90% of the pregnant women were HbAA in both hospitals respectively. The pregnant women with HbAA, in BMSH wee 180(90%) 48(24.0%) were infected, 20 were HbAS 7(3.5%) were infected. In BGH 162(81%) were HbAA, 60(30.0%) were infected and 38 were HbAS, 10(5.0%) had malaria parasitemia respectively. The result showed high infestation among HbAA as compared to HbAS.

Table. 4.5 Prevalence of malaria among pregnant women by Hb-Genotype.

	BMSH		BGH	
Hb-Genotype	**Number examined**	**Number positive**	**Number examined**	**Number positive**
AA	180(90)	43(24.0)	162(81)	60(30.00)
AS	20(10)	7(3.5)	38(19)	10(5.00)

Numbers in parenthesis=percentages.

DISCUSSIONS

The prevalence of malaria among pregnant women in BMSH was 27.5% and BGH was 35.0%, the prevalence of malaria was high among pregnant women attending antenatal clinic in BGH although statistically, there was no significant difference. In other Sub-Sahara Africa, the prevalence of malaria had been reported to be lower in urban than the rural areas (Baragalti *et al;* 2004). The factors contributing to this are, suitable breading site for mosquitoes, improper drainage systems, broken roads, pot holes, un-drained gutters, plantains planted beside dwelling houses which are good breeding sites for mosquitoes. Overcrowding which makes transmission very efficient and insecticide resistant vectors which make reduction in vector density impossible. Poverty can be implicated as it makes difficult the purchase of anti-malarial drugs, insecticides for residual indoor spaying and balanced diet for healthy living.

The prevalence of malaria by educational status showed that pregnant women who had just primary/secondary education had high prevalence of malaria in BGH (28.5%) as compared to (10.0%) for these with tertiary education. In BMSH, pregnant women who had tertiary education had high prevalence of malaria (17.5%), while pregnant women with primary/secondary education had (6.5%). The high prevalence in both hospitals corresponds to the two groups that majority of the pregnant women do not use INTs. Non use of ITNs plays a major role in the prevalence of malaria parasite infestation. The result obtained from BGH was in line with that of (Uneke, 2007) that women with none formal education had higher prevalence of malaria but this was not in agreement with the result from BMSH; both are influenced by the use of ITNs rather than educational status. Other incriminated factors that may be, cultural beliefs, poverty, poor understanding, nutritional status and occupation. The pregnant women whether educated or not, who failed to adhere to possible preventive measures especially, the use of INTs, had more infestation rate.

The results showed that there was significant difference at $P < 0.05$ between pregnant women with primary/secondary and tertiary education in BGH and also significant difference between women with tertiary education and primary/secondary in BMSH. This shows that the compliance whether absolute or partial has impact on the effectiveness of INTs. This was also noted by (Nyamngee *et al*; 2014). The use of INTs plays a key role in prevention and control of malaria in malaria endemic region such as the Sub-Sahara Africa. Over the last decade progress had been made in the control of malaria vectors in Sub-Sahara Africa, mainly using long lasting insecticidal nets (LLINs) and indoor residual spraying (IRS). Both preventive measures have greatly contributed to the decrease of malaria in Sub- Sahara Africa where the burden of malaria is heaviest (Barnes *et al*., 2017; Bhatt *et al*., 2017; WHO, 2016). The use of LLINs and IRS has placed a special advantage on *Anopheles species* that exhibit tendency to feed outdoor, early in the evening, during diurnal hours and on non humans. From this study, evidence strongly showed the reciprocal relationship between the use of ITNs or LLINs and the prevalence of malaria or malaria parasitemia in pregnancy as also noted by (Barnes *et al.,* 2017, Bhatt *et al.,* 2017). Massive education to impress the use INT and knowledge of the benefits of the using it, may enhance the use of INTs or LLNs (Onorride *et al*.,2015).

The prevalence of malaria among pregnant women by ABO blood group showed that, blood group O had high infestation rate compared to other blood groups as was also noted by (Pathirans *et al*; 2005; Zarihum *et al*, 2011; Amala and Nwibani, 2015). This result was not in line with finding of Uneke, 2007, Adajevwo, 2013 who concluded that all ABO blood groups were at equal chances of malaria parasite infestation. It had been established that rosetting is a virulent phenomenon of *Plasmodium falciperum* associated with severe malaria. *P. falcperum* encoded repetitive interspersed family of polypeptides (RIFINs) are expressed on the surface of infected red blood cells (RBCs) preferentially of group A to form large rosettes and mediate vascular binding of RBCs. RIFINs have fundamental role in development of severe malaria, thereby contributing to the varying global distribution of ABO blood group in human population (Sachi *et al*., 2015). RIFINs are adhesions implicated in severe *Plasmodium falciparum* malaria. In severe malaria associated with *P. falciperum* the infected RBCs adhere excessively in the micromusculature and block blood flow, causing oxygen deficiency and tissue damage which can lead to coma, brain damage and death. RIFINs makes it's way to the RBCs surfaces

where it acts as glue to bind strongly with the surface of blood group A, but only weakly to group O. This clearly shows other ABO blood groups aside O are exposed to more severe consequences.

In our findings, genotype HbAA had high prevalence of malaria compared to HbAS, the sickle cell trait, as soon as malaria parasite (*Plasmodiium falciparum)* begin to multiply in the RBCs, using the cell oxygen supply; the HbAS changes from round to sickle shape. The reduced oxygen level results in diminished parasite growth. Aside this, the malaria parasite cannot complete it's life cycle as a result of the sickle and destruction of infected RBCs; thereby preventing the progression of the disease (Amala and Nwigbani, 2015). The sickle cell trait produced higher level of superoxide oxygen ($O_{2)}$ and hydrogen peroxide (H_2O_2) which are toxic to a number of pathogens, including malaria parasite (Agarwal *et al*., 2000). HbAA is very susceptible to malaria parasite because the red blood cells are conducive to the growth and development of the parasites (Okwa, 2004).

CONCLUSIONS

The effective use of INTs or LLNs in the control *Anopheles* mosquitoes (vector) remains the gold standard measure to reduce the burden of malaria in Sub Sahara Africa; especially among pregnant women who bear more burden of malaria because of pregnancy situations.

REFERENCES

1. Agarwal A., Guindo A. Crsoko Y. Taylor J.G., Coulbay D., Kone A., Kayentae K., Djimde A. Plowe C.V. and Doumbo O. Haemoglobin C associated with protection from severe malaria in the Dogon of Mali, a West African population with low prevalence of haemoglobin S. Blood, 2000; 95: 2358-2363.
2. Amala, S.E. & Nwibani C.P. Malaria in pregnancy and it's association with ABO blood group and heamoglobin genotype. International Journal of Development Research, 2015; 5(8): 5317-3520.
3. Akligbe, R. E., Ige, S, F., Ndegunlola, G. J., Adewumi, M. O. & Azeez, M. O. Malaria Haemoglobin Genotypes and ABO Blood Groups in Ogbomoso, Nigeria. Journal of Tropical Medicine, 2011; 6(4): 73 – 76.
4. Bhatt S, Weiss DJ, Mappin B, Dalrymple U, Cameron E, Bisanzio D, et al. (2015) Coverage and system efficiencies of insecticide-treated nets in Africa from 2000 to 2017. eLife.. doi:10.7554/eLife.09672.
5. Barnes KG, Irving H, Chiumia M, Mzilahowa T, Coleman M, Hemingway J, et al. Restriction to gene flow is associated with changes in the molecular basis of pyrethroid resistance in the malaria vector Anopheles funestus. Proceeding of Natlonal Academy of Science, USA. 2017; 114: 286–91.
6. Cheesbrough, M. Measurement of Hemoglobin: in District Laboratory Practice in Tropical Countries. Part II: England, Cambridge University Press Ltd, 2000; 299-302.
7. Christine, M.C. and Walter, H.O. ABO blood group and Plasmodium falciperum. Blood, 2007; 110: 2251-2258.
8. Chin, J. Control of Communicable Diseases Manual. 17th ed. Washington: American Public Health Association, 2000; 20: 101-109.
9. Curtis, C., Maxwell, C., Lemnge, M., Kilama, W. L., Steketee, R. W., Hawley, W., Bergevin, Y., Campbell, C. C., Sachs J. & Snow, R. W. Scalling-up Coverage with Insecticide-Treated Nets against Malaria in Africa: who should pay? The Lancet Infectious Disease, 2003; 3: 304-307.
10. Guyatt, H. L. & Snow, R. W. Impact of Malaria During Pregnancy on Low Birth Weight in Sub-Saharan Africa. Clinical Microbiology Review, 2004; 17: 760-769.
11. Gallup, J. L. and Sachs, J. D. The Economic Burden of Malaria," American Journal of Tropical Medicine & Hygiene, 2001; 64(1-2): 85-96.
12. Greenwood, B. M., Bajang, K., Whitty, C. J. M. and Targett, G. A. T. (2005). Malaria. The Lancet, 23, 1487 – 1498.
13. Guyatt, H. L. & Snow, R. W. The epidemiology and burden of Plasmodium falciparum-Related Anaemia Among Pregnant Women in Sub-Saharan Africa, American Journal of Tropical Medicine & Hygiene, 2001; 64(1): 36 - 44.
14. Huddle, J. M., Gibson, R. S. & Cullinan, T. R. The Impact of Malaria Infection and Diet on the Anaemia Status of Rural Pregnant Malawian Women. European Journal of Clinical Nutrition, 1999; 53: 792 -801.
15. Kolawole, O. M., Babatunde, A. S., Jimoh, A. A. G., Balogun, O. R. & Kanu, I. G. Risk Determinants to Congenital Malaria in Ilorin, Nigeria. Asian Journal of Microbiology Biotechnical Environmental Science, 2006; 12(2): 215 – 222.
16. Mayagaya VS, Nkwengulila G, Lyimo IN, Kihonda J, Mtambala H, Ngonyani H, et al. The impact of livestock on the abundance, resting behavior and sporozoite rate of malaria vectors in southern Tanzania. Malaria Journal, 2015; 14-17.
17. Mayer, D. C., Kaneko, O., Hudson-Taylor, D. E., Reid, M.E. & Miller, L. H. Characterization of a Plasmodium falciparum Erythrocyte Binding Protein Paralogous to EBA-175. Proceedings of the National Academy of Sciences, 2001; 98: 5222-5227.
18. Menéndez, C., D'Alessandro, U. & Kuile, F. O. Reducing the Burden of Malaria in Pregnancy by Preventive Strategies. The Lancetof infectious disease, 2007; 7: 126-135.
19. Miller, L. H, Good, M. F. and Milon, G. Malaria pathogenesis. Science, 1994; 264: 1878 – 1883.
20. Odajevwo, F. D. Prevalence of Malaria Parasitamia and its Association with ABO blood Grouping Among Students of Igenedion University Okada, Nigeria. British Journal of Medicine and Medical Research, 2013; 3(4): 1164-1177.
21. Okwa, O. O. Preliminary investigations on malaria in sickle cell patients among pregnant women and

infants in Lagos Nigeria. Nig. J. Parasitol, 2004; 25: 81-85.
22. Onorride, E., Samson B.A., Omokhudu, I., Elijah, B.A. and Ernest, W. Determinants of the use of insecticide treated nets among pregnant women in Nigeria. International Journal of Women Health, 2017; 7: 665-661.
23. Richmond, A., Edmond, A., James, P., Ekene, K.N. and Johnson, N.B. (2016). Relative susceptibility of ABO groups to Plasmodium falciperum malaria in Ghana. Advances in Haematology Roland, B. and Silas L.S. Rhesus blood group system and malaria prevalence among students of the university of Dschang, Cameroon. Malaria World Journal, 2016; 7: 4.
24. Pathirans, S. L., Alles, H. K. & Bandara, S. ABO-Blood-Group Types and Protection Against Severe Plasmodium falciparum Malaria. Annals of Tropical Medical Parasitology, 2005; 99: 119 - 124.
25. Roll Back Malaria Partnership. The Contribution of Malaria Control to Maternal and New Born Health: A Pregnant Mother from Nigeria Embraces Malaria Prevention Progress and Impact Series, 2014; 10: 17.
26. Russell TL, Govella NJ, Azizi S, Drakeley CJ, Kachur SP, Killeen GF. (2011). Increased. proportions of outdoor feeding among residual malaria vector populations following increased use of insecticide-treated nets in rural Tanzania. Malaria Journal, 10: 80.
27. Sachi, G,. Mia, P., Kirsten, M., Nicholas, J., Lara, p., Akhouri, R.R. et al. RIFINs are adhesives implicated in severe Plasmodium falciparium malaria. Nature Medicine, 2015; 21: 314-317.
28. Sowunmi, A. & Odulola, A. M. Artemether Treatment of Recrudescent Plasmodium falciparum Malaria in Children. Tropical Medicine International Health, 1997; 2: 631–638.
29. Uneke, C. J. Congenital Plasmodium falciiperium malaria in Sub Saharan Africa. A rarity or frequent occurrence ? Parasitol. Res., 2007; 101: 835-842.
30. WHO/UNICEF/UNU (2001). Iron deficiency anaemia; assessment, prevention and control.
31. World Health Organization, Severe of falciparum malaria. Translation Royal Society Tropica Medicine and Hygiene, 2004; 94: 1-90.
32. World Health Organization (2015). Fact Sheet World Malaria Report 2015.htm.
33. WHO. World malaria report 2016. Geneva: World Health Organization; 2016 http://apps.who.int/iris/bitstream/10665/252038/1/9789241511711- eng.pdf?ua=1. Accessed 15 Feb 2017.
34. Zaruhum, T., Degarege, A. and Eko, B. Association of ABO blood group and Plasmodium malaria in Dora area South Ethiopia. Asian Journal of Tropical Biomedical Science, 2011; 1: 835-294.