

## Original Research Article

### **Variation in Some Haematological Parameters in Patients with Plasmodiasis attending Antenatal Clinic in Primary Health Care Centre, Ozuoba, Port Harcourt**

#### **ABSTRACT**

**INTRODOCTION:** Haematological parameters are useful in making diagnosis of diseases and also help in the antenatal assessment of women during pregnancy. The physiological changes of pregnancy, due to the influence of hormones such as oestrogen, progesterone also affect haematological indices when compared with the non-pregnant state.

**AIM:** The study investigated haematological indices in pregnant women with plasmodiasis attending antenatal clinics in Primary health care centre Ozuoba, Port Harcourt.

**METHODOLOGY:** This was a cross sectional study, which recruited a total of one hundred (100) subjects, of which (40) were non parasitized pregnant women who served as control, while (60) were parasitized pregnant women who served as test. The samples were analyzed for haematological parameters using (Sysmex XP-300), while microscopy was used to investigated for the presence of malaria parasite.

**RESULT:** Comparing the hematological parameters of the study population at different level of plasmodiasis, showed that WBC, and Neutrophil were significant with p-Value of 0.001 and 0.0089 respectively at  $P < 0.05$ .

The other haematological parameters such as, Red Blood Cell, Haemoglobin, Haematocrit, Mean Cell Volume, Mean Cell Haemoglobin, Mean CellHaemoglobin Concentration, Platelet, Plateletcrit, Neutrophils, lymphocyte, monocyte, eosinophil and basophil showed no statistical significant difference at  $p < 0.05$ . while comparison of the haematological parameters of the study group according to parity showed that WBC count had statistical significance with a p-Value of 0.0035 at  $p < 0.05$ .

**CONCLUSION:** Findings from this study has shown that there was statistical significant difference in the white blood cell count of malaria-parasitized pregnant subject. It was further revealed that there was no statistical significant difference in the other haematological parameters. The reason for the pattern of result obtained is that some of the pregnant women may have been receiving prophylactic antimalarial drugs before their recruitment, this will in turn minimize the impact of malaria infection on some haematological parameters.

**Keywords:** , Hamatological Parameters, Plasmodiasis, Pregnancy, Variation,

#### **Introduction**

Hematological parameters are useful in making diagnosis of diseases and also help in the antenatal assessment of women in pregnancy. The physiological changes of pregnancy, due to the influence of hormones such as oestrogen, progesterone, prolactin, renin, human placenta lactogen also affect haematological indices when compared with the non-pregnant state [1]. The haematological parameters is further influenced by race, geographical location, age, environmental factors and the prevalence of infectious diseases such as malaria [15]. On average, one in four pregnant women in areas of stable transmission in Africa has evidence of malaria

infection at the time of delivery based on the estimated prevalence of 26% of placental malaria[9].

In Sub-Saharan Africa, *Plasmodium falciparum* infection of the placenta remains a major challenge among pregnant women[2] and its prevalence is influenced by maternal age, gravidity, use of malarial prophylaxis, nutrition, host genetics, level of host's immunity, parasite genetics and transmission rates[5]. Maternal anaemia is the commonest consequence of *Plasmodium falciparum* malarial infection[2]. In Sub-Saharan Africa, Ataide *et al.* [3], in their study reported increase parasitaemia in the placenta is a risk factor for miscarriages in Pregnant women which in most cases may have adverse effect on the foetus. It is estimated that between 200,000 and 500,000 pregnant women develop severe anaemia as a result of malaria [Wessmar *et al.* (16). Anaemia is usually multifactorial in origin and although, malaria is an important contributor; nutritional deficiencies, helminth infection which is on the increase (>50% of the population) in malaria-endemic regions, HIV infections and genetic red blood cell disorders (Sickle cell and thalassemias) are other important contributing factors that is associated with the increase in malaria parasitaemia [10]. The hypersplenism in malaria infection is associated with a reduction in all three blood series that is, causing not only anaemia, but also thrombocytopaenia and leucopaenia [8]. According to WHO fact sheet and policy brief for the implementation of intermittent prevention of malaria in pregnancy, Malaria infection in pregnancy is considered to be risky, because of its adverse effect on the pregnant women, her foetus, and newborn, thereby creating serious public health concern [18,17]. Pregnant women are further subjected to stress as a result of infection with malaria parasites, most importantly, *P. falciparum* species.

Comment [B1]: Italicized

Comment [B2]: Recent works in malaria referenced

## Materials and Methods

### Study Design

This research work is a cross sectional study carried out among pregnant women who attended Ozuoba antenatal clinic. Questionnaires were administered to all the study subjects to determine their parities, social, occupational and marital status, including personal details. Blood samples were collected from study subjects who consented to participate in the study. Individuals who did not grant consent to participate in the study were excluded.

### **Study Population**

A total of one hundred (100) subjects were recruited for this study. Non parasitized pregnant women were used as control (40) while 60 parasitized pregnant women were used as test.

### **Study Area**

This study was conducted in Ozuoba Primary HealthCare Center, Port-Harcourt, Rivers State.

### **Eligibility of Participants**

#### **Inclusion Criteria**

Only antenatal patients having signs and symptoms of malaria visiting the above listed hospital were included in the research. Apparently healthy antenatal patients without signs and symptoms of malaria were used for the control.

#### **Exclusion Criteria**

Antenatal patients who are under drug were not involve in the research. Smokers and alcohol consumers were also excluded from the research.

### **Samples Collection**

Sample collection was done aseptically using the S-Monovette vacutainer blood collection system. After the tourniquet was applied 3-4 inches above the selected puncture site for not more than 1 minute, the selected puncture site was palpated with the gloved index finger and wiped with a wet swab. The vacutainer needle screwed into the holder was inserted into the lumen of the vein at 15-30 degree angle with the skin. The vacutainer anticoagulated tube (Ethylene diamine tetra acetate) was inserted into the holder and was filled by the blood up to 5ml. The tourniquet was removed first followed by the filled tube which was then mixed gently.

### **Methodology**

**Full Blood Count (FBC):** Measurement of haemoglobin, red blood cells, white blood cells and platelets count were done by automation using Haematology CBC auto analyzer SYSMEX KX-21N (Sysmex Corporation Japan, S/No B4 577), made by Beckman Coulter.

**Identification of Malaria Parasite by Giemsa Staining Technique:** On a well labeled clean grease-free-glass slide, a Pasteur pipette was used to pipette a certain quantity of the well-mixed sample. Then a drop of the sample was placed on the slide. A thick film was made by smearing the blood on the slide. Thereafter, the film was allowed to air dry. The slide was then flooded with a 1 in 30 dilution of Giemsa stain for 30 minutes. After staining, the slide was rinsed in water and blotted dry with cotton wool. The slide was allowed to air dry. After drying, a drop of immersion oil was placed on the stained portion of the slide and viewed under the light microscope using 40x and 100x objective lenses to focus and view respectively with the iris diaphragm opened and condenser rack up.

#### **Interpretation of Results**

1-10/10HPF = +++

10-20/10HPF = ++++

1-10/100HPF = +

11-20/100HPF = ++

#### **Data Analysis**

The data generated were analyzed using GraphPad Prism (version 8.02) Comparison of some haematological indices between antenatal patients with signs and symptoms of malaria and statistical tools such as student's t-test and one way ANOVA were used. Results were presented as mean  $\pm$  SD with statistical significance set at  $p < 0.05$ .

**Ethical Approval:** Ethical consent was sort and gotten for this research.

#### **Results**

Table 1 shows the Demographic Characteristics of Malaria Parasitized Subjects. Age groups of the population ranged from 19-25, 26-32, 33-40 with the frequency distribution of the 0.17, 0.63 and 0.20 and percentage of 17%, 63% and 20% respectively.

The educational status of the study population showed that SSCE, Tertiary and post graduate had frequency distribution of 0.55, 0.35 and 0.10 and percentage of 55%, 35% and 10% respectively.

The parity of the study population ranged from 0-1, 2-3, and 4-5 with the frequency distribution of 0.57, 0.37 and 0.06 respectively and the percentage of 57%, 37% and 6% respectively.

The occupation of participants in this study included business, teaching and student with the frequency distribution of 0.55, 0.40 and 0.05 and percentage of 55%, 40% and 5% respectively.

**Table 1 Demographic Characteristics of Malaria Parasitized Subjects**

<b>Subjects</b>	<b>No. of Participants</b>	<b>Frequency</b>	<b>Percentage</b>
<b>Age Groups</b>			
19-25	10	0.17	17%
26-32	38	0.63	63%
33-40	11	0.20	20%
<b>Education Status</b>			
SSCE	33	0.55	55%
Tertiary	21	0.35	25%
PostGrad.	6	0.10	10%
<b>Parity</b>			
0-1	34	0.57	57%
2-3	22	0.37	37%
4-5	4	0.06	6%
<b>Occupation</b>			
Business	33	0.55	55%
Teaching	24	0.40	40%
Students	3	0.05	5%
<b>Level of Malaria</b>			
+	43	0.72	72%
++	17	0.28	28%

Table 2 shows the Hematological Parameters of Malaria Parasitized Subjects and Non-Parasitized Subjects. Comparing the hematological parameters of the study group. It was seen that WBC had a Mean  $\pm$  SD of  $8.73 \pm 2.27$  and  $6.89 \pm 1.51$  for the test and control respectively. There was a statistical significant difference with P- Value of 0.0036 at  $P < 0.05$ . Further comparison of the test and the control subjects showed no statistical significant difference at  $p < 0.05$  for Red Blood Cell (RBC), Haemoglobin (HB), Haematocrit (HCT), Mean Cell Volume (MCV) Mean Cell Haemoglobin (MCH), Mean Cell Haemoglobin Concentration (MCHC), Platelet (PLT), Plateletcrit (PCT), Neutrophil, lymphocyte, monocyte, eosinophil and basophil.

**Table 2: Comparative Analysis of Haematological Parameters of Malaria Parasitized Subjects Against Non-Parasitized Subjects**

Parameters	Malaria Parasitized Subjects (Test)	Non- Parasitized Subjects (Control)	pvalue	fvalue	Remark
WBC( $\times 10^9/L$ )	$8.73 \pm 2.27$	$6.89 \pm 1.51$	0.0036	3.015	S
RBC ( $\times 10^9/L$ )	$3.88 \pm 0.52$	$3.78 \pm 0.36$	0.4878	0.697	NS
HB (g/dl)	$10.85 \pm 1.06$	$11.06 \pm 0.94$	0.4788	0.712	NS
HCT (%)	$32.02 \pm 3.62$	$32.69 \pm 3.17$	0.5058	0.664	NS
MCV (fL)	$82.98 \pm 6.86$	$86.51 \pm 5.59$	0.0654	1.874	NS
MCH(pg)	$28.17 \pm 2.77$	$29.30 \pm 2.23$	0.1405	1.491	NS
MCHC(g/dl)	$262.9 \pm 139.7$	$267.9 \pm 141.0$	0.9007	0.125	NS
PLT ( $\times 10^9/L$ )	$197.2 \pm 60.23$	$176.4 \pm 44.67$	0.2046	1.281	NS
RDW-SD (%)	$44.55 \pm 5.28$	$42.82 \pm 10.99$	0.3872	0.870	NS
RDW-CV (fl)	$14.27 \pm 2.23$	$13.76 \pm 1.22$	0.3849	0.874	NS
PDW	$14.92 \pm 2.31$	$15.06 \pm 2.21$	0.2156	0.829	NS
MPV (%)	$10.58 \pm 1.07$	$10.73 \pm 0.77$	0.6020	0.524	NS
P-LCR	$55.37 \pm 23.53$	$49.13 \pm 18.93$	0.3371	0.966	NS
PCT (mL/L)	$1.67 \pm 1.00$	$1.425 \pm 0.84$	0.3681	0.904	NS
NEUT( $\times 10^9/L$ )	$52.64 \pm 28.45$	$51.22 \pm 27.87$	0.8606	0.176	NS
LYM( $\times 10^9/L$ )	$18.49 \pm 11.19$	$20.22 \pm 11.76$	0.5944	0.176	NS
Mono( $\times 10^9/L$ )	$3.69 \pm 2.11$	$4.18 \pm 2.52$	0.4387	0.779	NS
Eosin( $\times 10^9/L$ )	$1.10 \pm 1.25$	$1.22 \pm 1.26$	0.7380	0.335	NS
Baso ( $\times 10^9/L$ )	$0.03 \pm 0.06$	$0.04 \pm 0.07$	0.9273	0.091	NS

. keys: S=Significant, NS=Not Significant.

Table 3 shows the Hematological Parameters of Malaria Non-Parasitized Subjects and Malaria Parasitized Subjects at Different Level of Plasmodiasis Quantification. Comparing the hematological parameters at different level of plasmodiasis, it was seen that white Blood cell (WBC), and Neutrophil showed significance with p-Value of 0.001 and 0.0089 respectively at  $P<0.05$ . Comparison of other parameters such as Red Blood Cell (RBC), Haemoglobin (HB), Haematocrit (HCT), Mean Cell Volume (MCV) Mean Cell Haemoglobin (MCH), Mean Cell Haemoglobin Concentration (MCHC), Red cell Distribution Width-Standard Deviation (RDW-SD), Red cell Distribution Width-Coefficient Variation (RDW-CV), Platelet Distribution Width (PDW), Mean Platelet Volume (MPV), Platelet-Larger Cell Ratio (PLT-LCR), Plateletcrit (PCT), lymphocyte, monocyte, eosinophil and basophil showed no statistical significance difference at  $P<0.05$ .

**Table 3: One-WAY ANOVA of Haematological Parameters of Malaria Non-Parasitized Subjects (control) against Malaria Parasitized Subjects (Test) at Different Level of Plasmodiasis Quantification**

parameters	Control	Malaria (+)	Malaria (++)	pvalue	fvalue	Remark
WBC( $\times 10^9/L$ )	6.897 $\pm$ 1.519 <sup>a</sup>	8.138 $\pm$ 1.922	10.56 $\pm$ 2.390 <sup>b</sup>	<0.001	13.17	S
RBC ( $\times 10^9/L$ )	3.787 $\pm$ 0.3686	3.914 $\pm$ 0.5633	3.798 $\pm$ 0.4107	0.6054	0.5058	NS
HB (g/dl)	11.06 $\pm$ 0.9473	10.86 $\pm$ 1.150	10.79 $\pm$ 0.7740	0.7626	0.2722	NS
HCT (%)	32.69 $\pm$ 3.176	32.17 $\pm$ 3.931	31.58 $\pm$ 2.537	0.7054	0.3509	NS
MCV (fL)	86.51 $\pm$ 5.594	82.74 $\pm$ 6.695	83.73 $\pm$ 7.594	0.1664	1.843	NS
MCH(pg)	29.30 $\pm$ 2.233	28.00 $\pm$ 2.753	28.69 $\pm$ 2.878	0.2444	1.439	NS
MCHC(g/dl)	267.9 $\pm$ 141.0	259.0 $\pm$ 141.7	275.0 $\pm$ 138.3	0.9321	0.070	NS
PLT ( $\times 10^9/L$ )	176.4 $\pm$ 44.67	201.3 $\pm$ 63.51	184.8 $\pm$ 48.91	0.3010	1.223	NS
RDW-SD (%)	42.82 $\pm$ 10.99	45.23 $\pm$ 5.611	42.48 $\pm$ 3.560	0.3226	1.151	NS
RDW-CV (fl)	13.76 $\pm$ 1.229	14.52 $\pm$ 2.460	13.48 $\pm$ 1.020	0.1947	1.677	NS
PDW	15.06 $\pm$ 2.214	14.78 $\pm$ 2.346	15.34 $\pm$ 2.257	0.7357	0.3083	NS
MPV (%)	10.73 $\pm$ 0.7795	10.51 $\pm$ 1.072	10.79 $\pm$ 1.109	0.5987s	0.5170	NS
P-LCR	49.13 $\pm$ 18.93	55.00 $\pm$ 23.77	56.48 $\pm$ 23.72	0.6202	0.4812	NS
PCT (mL/L)	1.425 $\pm$ 0.8475	6.478 $\pm$ 30.01	1.660 $\pm$ 0.9782	0.6808	0.3867	NS
NEUT( $\times 10^9/L$ )	64.32 $\pm$ 6.704 <sup>a</sup>	69.84 $\pm$ 4.986 <sup>b</sup>	68.99 $\pm$ 7.366	0.0089	5.082	S

LYM( $\times 10^9/L$ )	26.14 $\pm$ 8.186	24.06 $\pm$ 4.613	24.93 $\pm$ 6.703	0.5022	0.6960	NS
Mono( $\times 10^9/L$ )	4.183 $\pm$ 2.525	4.840 $\pm$ 1.106	4.308 $\pm$ 1.274	0.2918	1.255	NS
Eosin( $\times 10^9/L$ )	1.229 $\pm$ 1.267	1.245 $\pm$ 1.168	1.769 $\pm$ 1.246	0.3678	1.015	NS
Baso( $\times 10^9/L$ )	0.03 $\pm$ 0.02	0.01 $\pm$ 0.005	0.03077 $\pm$ 0.07	0.2477	1.426	NS

Post-Hoc Analysis: WBC & NEUT: Values in the same row with different superscripts (a, b) differ from

each other at  $p < 0.05$ . Keys: S=Significant, NS=Not Significant.

## Discussion

This research work was a cross sectional study carried out among pregnant women who attended Primary Health Care Centre, Ozuoba antenatal clinic, Port Harcourt Rivers State

Assessing the demographic distribution of the study population, it was shown that the age groups of the population ranged from 19-25, 26-32, 33-40 with the frequency distribution of the 0.17, 0.63 and 0.20 representing 17%, 63% and 20% respectively. This is in contrast to a study by [14], in their study they observed that younger women in the age group 21-25 years constituted a significant number of the subjects(36.7%) used in their study.

Majority of the malaria parasitized subjects had only Secondary education(55%), this was followed by those who attained tertiary level of education (35%) then followed by women with post graduate education(10%).This finding is in agreement with a recent report by Olusegun-Joseph *et al.* (12),They reported increased susceptibility to malaria parasite on women of Ijede community of Lagos state, Nigeria who were mostly traders with little or no knowledge on malaria prevention strategies, and not well educated. In another study by Hemingway *et al.* [6], they reported low parasitaemia on individuals who had higher educational exposure, knowledge of malaria prevention strategies, consistent drainage cleaning and proper waste disposal. This is suggestive of the fact that the level of education can play a role in preventing malaria infection. High standard of education usually affect health awareness and therefore has a positive impact on healthy living and lifestyle.

Comparing the hematological parameters of the study group. It was seen that WBC had a statistical significant difference with a p-Value of 0.0036 at  $p < 0.05$ . The other hematological parameters such as Red Blood Cell (RBC), Haemoglobin (HB), Haematocrit (HCT), Mean Cell



Volume (MCV), Mean Cell Haemoglobin (MCH), Mean Cell Haemoglobin Concentration (MCHC), Platelet (PLT), Plateletcrit (PCT), Neutrophil, lymphocyte, monocyte, eosinophil and basophil showed no statistically significant difference at  $P < 0.05$ .

This is in agreement with a study by Bakhubaira (4) and Osaro *et al.* [13] who reported that WBC was elevated in malaria-parasitized pregnant women. This may be due to the fact that white blood cells are responsible for the body defense during pregnancy. However, the findings in this study is in contrast with a report by Maina *et al.* (7), who reported that malaria parasitemia have no effect on all the hematological parameters when 500 pregnant women in Kano were assessed based on parity. This disparity in the findings may arise as a result of some pregnant women in their study, probably being on prophylactic antimalarial drugs before their recruitment, thus causing a reduced effect of malaria parasitemia on other haematological parameters.

Comparing the hematological parameters of the study population at different level of plasmodiasis, it was seen that WBC, and Neutrophil were significant with p-Value of 0.001 and 0.0089 respectively at  $P < 0.05$ . Comparison of other parameters such as Red Blood Cell (RBC), Haemoglobin (Hb), Haemoglobin (HCT), Mean Cell Volume (MCV), Mean Cell Haemoglobin (MCH), Mean Cell Haemoglobin Concentration (MCHC), Red Cell Distribution Width-Standard Deviation (RDW-SD), Red Cell Distribution Width-Coefficient Variation (RDW-CV), Platelet Distribution Width (PDW), Mean Platelet Volume (MPV), Platelet- Large Cell Ratio (P-LCR), Plateletcrit (PCT), lymphocyte, monocyte, eosinophil and basophil showed no statistical significant difference at  $P < 0.05$ . This is in agreement with a study by Muwong *et al.* [11]. In their study, it was seen that increase in plasmodiasis is directly proportional to WBC.

Comment [B3]: Abbreviation defined

## Conclusion

Findings from this study has shown that there was a significant difference in white blood cell count of malaria-parasitized pregnant subjects. It further revealed that there was no statistical significant difference in the other haematological parameters.

## References

**Comment [B4]:** Vancouver system of references applied

1. Adnan A. The role of laboratory medicine for health during pregnancy. *Journal of International Federation of Clinical Chemistry and Laboratory Medicine*. 2018; 29: 280-84 .
2. Aguzie ION. Pregnancy-associated Malaria, Challenges and Prospects in Sub- Saharan Africa. *Clinics in Mother Child Health*. 2018; 15: 282.
3. Ataide R, Mayor A, Rogerson SJ. Malaria, primigravidae, and antibodies: knowledge gained and future perspectives. *Trends in Parasitology*. 2014; 30: 85–94.
4. Bakhubaira S. Hematological parameters in severe complicated *Plasmodium falciparum* malaria among adults in Aden. *Turkish Journal of Haematology*. 2013; 30: 394-99.
5. Gething PW, Casey DC, Weiss DJ, Bisanzio D, Bhatt S, Cameron E. *et al*. Mapping *Plasmodium falciparum* Mortality in Africa between 1990 and 2015. *New England Journal of Medicine*. 2016; 375(25): 2435-45.
6. Hemingway J, Shretta R, Wells TN, Bell D, Djimdé AA, Achee N, Qi G. Tools and strategies for malaria control and elimination: what do we need to achieve a grand convergence in malaria? *PLoS biology*. 2016;14(3): 1.
7. Maina RN, Walsh D, Gaddy C, Hongo G, Waitumbi J, Otieno L. *et al*: Impact of *Plasmodium falciparum* infection on haematological parameters in children living in Western Kenya. *Malaria Journal*. 2010; 9 (3): 4-10.
8. Markus MB. Malaria eradication and the hidden parasite reservoir. *Trends in parasitology*. 2017; 33(7): 492-95.
9. Mayor A, Bardaji A, Macete E, Nhampossa T, Fonseca AM, Gonzalez R. *et al*. . Changing trends in *P. falciparum* burden, immunity, and disease in pregnancy. *New England Journal of Medicine*. 2015; 373: 1607–17.
10. Mulu A, Legesse M, Erko B, Belyhun Y, Nugussie D, Shimelis T, *et al*. Epidemiological and clinical correlates of malaria-helminth co-infections in Southern Ethiopia. *Malaria Journal*. 2013;12:227.
11. Muwonge H, Kikomeko S, Sembajjwe LF, Seguya A, Namugwanya C. How reliable are hematological parameters in predicting uncomplicated *Plasmodium falciparum* malaria in an endemic region?. *International Scholarly Research Notices*. 2013; 2(5): 36-55.
12. Olusegun-Joseph T, Oboh M, Ovioma G, Fagbohun I, Okorafor U, Aina D. Differential Prevalence of Malaria Infection in Rural and Urban Out-Patient Clinics in Lagos State, Nigeria. *Pan African Journal of Life Sciences*. 2019; 2: 79-84.

13. Osaro E, Abdulirahaman A, Erhabor T. Effect of malaria parasitaemia on some haematological parameters of pregnant women of African Descendants in specialist hospital, Sokoto, North Western Nigeria. *JOJ Nursing and Health Care*. 2019; 10(4); 555795.
14. Panti AA, Omokanye LO, Ekele BA, Jiya NMA, Isah AY, Nwobodo EI. et al. The prevalence of asymptomatic malaria parasitaemia at delivery in usmanudanfodiyo university teaching Hospital Sokoto North western Nigeria. *Global Resource Journal*. 2012; 2(9): 48-53.
15. Purohit G, Shah T, Harsoda JM. Hematological profile of normal pregnant women in Western India. *Scholars Journal of Applied Medical Science*. 2015; 3:2195-99.
16. Wassmer SC, Taylor TE, Rathod PK, Mishra SK, Mohanty S, Arevalo-Herrera . Investigating the Pathogenesis of Severe Malaria: A Multidisciplinary and Cross-Geographical Approach. *American Journal of Tropical Medicine and Hygiene*. 2015; 93(3): 42-56.
17. World Health Organization ,2016; Fact sheet: World Malaria Report 2015.
18. World Health Organisation WHO policy brief for the implementation of intermittent preventive of malaria in pregnancy using sulfadoxin-pyrimethamine (IPTp-SP). WHO Global Malaria Programme. 2014.