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# Influence of Age Category and Malaria Type on Some Haematological, VWF, ADAMTS13, L-arg, D2D, FBG, FVIII and Coagulatory Parameters of Malaria Infected Paediatric Subjects in Rivers State, Nigeria

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***Authors' contributions***

*This work was carried out in collaboration among all authors. Author ZAJ designed the study. Author EME wrote the protocol. Author NCI wrote the first draft of the manuscript. Authors BSM and SOA managed the analyses of the study and the literature searches. All authors read and approved the final manuscript.*

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## ABSTRACT

**Aim:** The aim of the study was to assess the influence of age category and malaria type on haematological and haemostatic parameters in malaria infected children resident in Rivers State, Nigeria.
**Study Design:** The study was cross-sectional observational study.
**Place and Duration of Study:** University of Port Harcourt Teaching Hospital, Rivers State, Nigeria, between the month of March and August 2020.
**Methodology:** A total of 822 pediatrics (0-16 years), were randomly selected for this study after due parental consent. 5ml of venous blood was collected from each subject: 1ml was dispensed into paediatric EDTA (for haematologic and parasite density) and 4ml into sodium citrate bottle for L-arginine assay by ELISA-method, while Full blood count was determined using haematological

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auto-analyser, Mindray BC-6800. Malaria density was determined by microscopic method using thick and thin Giemsa stained blood smears. Level of significance was set at P<0.05.

**Results:** The results showed that there were significant (*p<.001)* decreased in WBC & NEU in neonate and adolescent and increased in children when compared with the control subjects of uncomplicated and complicated malaria. EOS was observed to have a significant (p< 0.0065) increased in the age categories; (neonates, children and adolescent) however complicated malaria subjects were observed to have a significant (p<.05) decreased EOS when compared with the uncomplicated malaria. In addition, there was no significant (p>0.05) different in the mean PT, PTT, INR, ADAMT13, and FViii in the three age categories. However, FBG, VWF and D2D were observed to be significantly (*p<.05*), (*p<.05)*, (*p<.001)* increased in the age categories being highest in children.

**Conclusion:** In conclusion, haematological parameters decreased as malaria got complicated in neonates, increased in children, while haemostatic parameters such as VWF, FBG and D2D increased in the three age categories being highest in children.

*Keywords: Age category; malaria; haematologic and haemostatic parameters; paediatric; Rivers State; Nigeria.*

## 1. INTRODUCTION

Malaria remains a significant global health challenge, with infections causing disease symptoms that range from uncomplicated to complicated cases. In spite of a declining trend in the number of cases and deaths over the last few years, malaria still causes significant mortality and morbidity and the bulk of the burden is observed in children under 5 years of age [1]. Controversy exists as to whether a coagulopathy is primarily or partially involved in the pathogenesis of malaria [2] for a number of reasons: (i) typical bleeding is not present in most *P. falciparum*-infected patients [3,2] and occurs in 5% to 10% of severe malaria cases (1% of all cases), although clinically not significant, can be confirmed as a compensated state according to *in vivo* coagulation tests [4] (ii) thrombocytopenia in *falciparum* malaria was indicative of prognosis in one study (iii) fibrin indicative of *in vivo* activation of the coagulation cascade have been clearly reported in some studies but infrequently found or undetectable in others could be explained by the fact that coagulation activation is accompanied by compensatory fibrinolysis [5,6].

In African children with *P. falciparum* malaria, von Willebrand factor (VWF) levels were associated with severity, with the highest levels being observed in complicated malaria (CM) and uncomplicated malaria [1]. VWF is a large multimeric glycoprotein involved both in platelet adhesion and aggregation, and is produced predominately by activated endothelial cells. When released, VWF is in an activated conformation, which allows interaction with the platelet receptor gpIa/V/IV and triggers intravascular platelet aggregation [7]. This function primarily occurs under high shear stress conditions on the arteriolar side of the microcirculation [8]. One possible mechanism for increased VWF levels might be decreased activity of the VWF-cleaving protease ADAMTS13 (a disintegrin and metalloproteinase with a thrombospondin type 1 motif, member 13) and ADAMTS13 activity has been shown to be reduced in other conditions associated with thrombocytopenia and raised VWF levels, such as bacterial sepsis and thrombotic thrombocytopenic purpura [7].

However in early experimental malaria, raised VWF and thrombocytopenia were not accompanied by a change in ADAMTS13 activity [9]. These results suggest that a different mechanism for increased vWF levels may be implicated in malaria. The release of large multimers of VWF and adherent platelets into the circulation might provide multiple binding sites for PfIE and could potentially lead to major rheological disturbances by generating circulating clumps of VWF, platelets and PfIE. The possible association between an increase in VWF and mortality still remains to be investigated in adult and paediatric malaria severity. *In-vitro*, endothelial nitric oxide (NO) decreases tissue factor (TF) expression [10] and is the major inhibitor of the exocytosis of Weibel-Palade bodies. It has recently been shown that endothelial NO is reduced in severe malaria in adults and is associated with increased concentrations of angiopoietin-2, an angiogenic factor also stored in Weibel-Palade bodies [11].

Haematological parameters are measurable indices of the blood that serve as a marker for disease diagnosis. Haematological changes are some of the most common complications in malaria and they play a major role in malaria pathogenesis. Also, changes in these haematological parameters are likely to be influenced by any disease condition including endemic diseases, such as malaria, G6PD deficiency that can affect health of human with various clinical presentations. The influence in haematological parameters by any disease shows that haematological changes are reliable laboratory indicators of malaria *Plasmodium falciparum.* These changes involve the complete full blood count such as leucocytes, red blood cells and thrombocytes [12].

Malaria severity has been associated with thrombocytopenia and considered an independent predictor of death, including in *falciparum* malaria [6] and vivax malaria [13]. However, there are other reports showing that there is no direct link between the level of thrombocytopenia and the severity of malaria [14,15]. Severe malaria is rare in early infancy [16]. In endemic areas neonate may have cord blood and peripheral parasitaemia which usually disappears within hours or days. Illness in the neonate resulting from malarial infection is uncommon, but may present as fever, anaemia and probably neonatal jaundice few days after delivery. Over the next few months of life, parasitaemia appears in an increasing proportion of children. The greater majority of infections is symptomless and is reflected in the high prevalence of asymptomatic parasitaemia among children in endemic areas [16]. Therefore, the aim of the study was to assess the influence of age category and malaria type on haematologicl and haemostatic parameters in malaria infected children resident in Rivers State, Nigeria.

## 2. MATERIALS AND METHODS

### 2.1 Study Area

The study was carried out in Port Harcourt, the capital city of Rivers State Nigeria. Port Harcourt is situated within geographical co-ordinates 4°′49′27″N 7°2′1″E. Port Harcourt features tropical wet climates with lengthy and heavy Rainy seasons and very short dry seasons. Only the months of December and January truly qualifies as dry season months in the city. The harmattan, which climatically influences many cities in West Africa, is less pronounced in Port Harcourt. Port Harcourt's heaviest precipitation occurs during September with an average of 367 mm of rain. December on average is the driest month of the year with an average rainfall of 20 mm. Temperatures throughout the year in the city are relatively constant, showing little variation throughout the course of the year. Average temperatures are typically between 25°C-28°C in the city. University of Port Harcourt Teaching Hospital, Rivers State, Nigeria; was used as the Centre for the study. University of Port Harcourt Teaching Hospital, Choba Port Harcourt was established in 1980 with 500 bed spaces.

### 2.2 Study Population

A total of eight hundred and twenty two (822) paediatrics with age range of 0-16 years were randomly selected. Children registered in the paediatric ward of University of Port Harcourt Teaching Hospital, UPTH with suspected *P. falciparum* parasitaemia presenting with complicated and uncomplicated febrile illness and children without febrile illness for immunization attending accident and emergency unit and the clinic were recruited for this study after obtaining a written consent from the parents or guardians from each participant. Their demographical information was collected using a questionnaire. The subjects were divided into three groups based on the clinical manifestations of malaria classified according to the definitions and associated criteria by the World Health Organization [17]. The uncomplicated malaria group with mild to moderate symptoms characterize by fever and lack of severe malaria. The complicated malaria group was defined by one of the WHO criteria as one with severe high parasitaemia (≥ 100,000 parasites/μL) [17], and apparently healthy age matched children without parasitological evidence of malaria.

### 2.3 Sample Size

Randomized method was used in the selection of subjects, taking into consideration, the total number of patient registered in the paediatrics clinic in University of Port Harcourt Teaching Hospital, Rivers State. The optimum sample size was obtained using the prevalence of malaria in children in Port Harcourt as 60.6% [14] and the sample size was calculated using the Cochran's sample size formula as shown below [18].

$$N = \frac{Z^2 pq}{d^2}$$

Where

N = The desired sample size
Z = The Standard Normal deviate usually set at 1.96 corresponding to the 95 confidence level
p = The prevalence of target population (60.6%)
q = 1- p
d = degree of accuracy desired set at 0.05

Therefore,

$N = ((1.96)^2 \times 0.606 \times (1-0.606)) / (0.05)^2$

N = 366.894

By adding 10% of non-respondent, the sample size was 404.

## 2.4 Inclusion and Exclusion Criteria

Children with and without fever of both sexes who were out-patient and/or admitted in the ward of University of Port Harcourt Teaching Hospital with aged range 0-16 years were included in the research with informed consent obtained from the parent/guardian of each participant. Whilst, children above 16 years of age with or without fever of both sexes who were not registered in the paediatric unit of University of Port Harcourt Teaching Hospital were excluded from the research.

## 2.5 Sample Collection and Handling

All recruited subjects were given study numbers which was used for all data collection /laboratory processes. 5ml of venous blood was withdrawn with minimum stasis under aseptic conditions from the dorsum of the hand or ante-cubital vein as the case may be [19] and dispensed into EDTA (1ml) and 4ml into sodium citrate anticoagulated tube. The sample bottles were then assigned a study code with a non-water soluble ink with date, sex and time of collection and logged on to a paper log after dispensing the blood sample into the sample bottles. The sample was rocked gently to mix and kept at room temperature and then analyzed within 4 hours of samples collection. Malaria parasites slide were made within 1 hour of collection to prevent loss of morphological characters of the *Plasmodium falciparum.*

## 2.6 Design of the Study

The study was a cross sectional study carried out on 822 paediatric patients with suspected malaria infection and children for immunization as control registered in university of Port Harcourt Teaching Hospital, Rivers state Nigeria from the month of March to August 2020. Following administrative clearance and ethical approval for the study, informed consent forms explaining the purpose, risks, benefit of the study and the volume of blood to be collected from each child were given to parent or guardian. Emphasis was laid on the voluntary participation of the children in the study and on the point that their refusal to participate in the study will in no way affect the treatment quality the child was to receive. Only those who signed the consent forms were enrolled into the study. The parents or guardians were free at any point in time to stop the participation of the child in the study. The average ages of the patients with mild malaria, severe malaria and non-malaria infected were ranged 0-16 years. The demographical data of the subjects were obtained using a questionnaire as well as their clinical manifestations after obtaining consent from the participant prior to blood sample collection. These included age, sex, fever (≥$37.5^0$c using digital thermometer by the medical personnel in charge) vomiting, dizziness, respiratory distress, diarrhea and coma. Blood sample was collected from each child for the determination of malaria parasite status, full blood count evaluation and coagulation parameters. The subjects were divided into three groups based on the clinical manifestations of malaria classified according to the definitions and associated criteria by the World Health Organization [17]. The complicated malaria group was defined by one of the WHO criteria as one with severe high parasitaemia (≥ 100,000 parasites/μL) [17]. Otherwise, it was considered uncomplicated malaria, and apparently healthy age matched children without parasitological evidence of malaria. Malaria parasite density, ADAMTS13, VWF, FVIII, L-arginine, D-dimer, Fibrinogen, PT, INR, PTT and white blood cells differentials were determined in these three groups and comparison made.

## 2.7 Methods of Assay

### 2.7.1 Malaria diagnosis

Light microscopy of thick and thin Giemsa stained blood smears method was used for diagnosing malaria as described by [20]. These stains contain eosin which is an acidic anionic dye and methylene blue (azure) which are basic cationic dyes. When diluted in buffered water at pH of 7.2, ionization occurs. Eosin component

stains the parasite nucleus red, while the methylene blue components stain the cytoplasm blue.

### 2.7.2 Determination of haematological indices

Haematology indices were analyzed using Mindray BC-6800, an auto Haematology analyzer system [21]. This is based on a combination of light scatter, electrical impedance, fluorescence, light absorption, and electrical conductivity methods to produce complete red blood cell, platelet, and leukocyte analyses.

#### *2.7.2.1 Quality control of full blood count determination*

The use of controls and a proper dilution of the sample before analysis were ensured to prevent too high number of events passing through the laser and ensure a better accuracy of the readings. Also, there is an automatic in built control programme in the auto analyzer. When the power button on the main unit was turned on, the main unit entered an auto rinse mode in which the instrument was rinsed three to five times, waste was drained, background counts were reviewed and the electronics, injector piston motors, bubble memory, and instrument status were automatically checked. (An auto rinse switch is available to the operator for a background check whenever desired). Manually prepared blood films were viewed for clot formation for any substantial abnormal value.

### 2.7.3 Determination of L-arginine: L-arginine activity and D-dimer (D2D)

L-Arginine: L-arginine activity and D2D was determined using Bioassay Technology Laboratory enzyme linked immunosorbent (ELISA) kit and Labtech Auto ELISA Plate Reader for L-arginine and D2D Activity. This assay employs the inhibition enzyme immunoassay technique. The microliter plate is pre-coated with Arginine (Arg) protein. Standards or samples are then added to the appropriate microliter plate wells with a biotin-conjugated antibody specific to Arginine (Arg). Next, Avidin conjugated to Horseradish Peroxidase (HRP) is added to each microplate well and incubated. After TMB substrate solution is added. The enzyme-substrate reaction is terminated by the addition of sulphuric acid solution and the color change is measured spectrophotometrically at a wavelength of 450nm±10nm. The concentration of Arginine (Arg) in the samples is then determined by comparing the optical density (OD) of the samples to the standard curve.

### 2.7.4 Estimation of some haemostatic parameters

#### *2.7.4.1 Prothrombin Time (PT)*

Prothrombin Time was analyzed manually using fortress PT reagent and Uniscope SM801A Laboratory Water Bath for précised temperature control throughout the range [22]. The calcium in whole blood is bound by sodium citrate, thus preventing coagulation. Tissue Thromboplastin, to which calcium has been added, is mixed with the plasma, and the clotting time is noted.

#### *2.7.4.2 Activated Partial Thromboplastin Time (APTT)*

Activated Partial Thromboplatin Time was analysed manually using fortress APPT reagent and Uniscope SM801A Laboratory Water Bath for précised temperature control throughout the range [22].

#### *2.7.4.3 ADAMTS13 Activity Assay*

ADAMTS13 activity was determined using Bioassay Technology Laboratory ELISA kit and Labtech Auto ELISA Plate Reader [23].

#### *2.7.4.4 Von Willibrand factor activity assay*

Von Willibrand Factor (vWF) Activity was determined using Bioassay Technology Laboratory enzyme linked immunosorbent (ELISA) kit and Labtech Auto ELISA Plate Reader [24].

#### *2.7.4.5 Fviii activity assay*

Factor VIII (Fviii) activity was determined using Bioassay Technology Laboratory enzyme linked immunosorbent (ELISA) kit and Labtech Auto ELISA Plate Reader [24].

## 2.8 Statistical Analysis

Statistical Analysis System (SAS) 9.4 was used to analyze data generated. Testing differences and comparison between means was done using One-way Analysis of variance (ANOVA). Variable such as age was presented as percentages. Post hoc test was conducted using the Tukey's Honestly Significant Difference (HSD) to ascertain the difference between the subjects. Level of significance was set as $P<0.05$.

**Table 3.1. Influence of age category and malaria type on haematological parameters (WBC, NEU, LYM, MON, EOS and BAS) of malaria infected paediatric subjects**

| Interactive Measures | | N | WBC ($10^9$/L) | NEU ($10^9$/L) | LYM ($10^9$/L) | MON ($10^9$/L) | EOS ($10^9$/L) | BAS ($10^9$/L) |
|---|---|---|---|---|---|---|---|---|
| **Age Category** | **Malaria Type** | | | | | | | |
| Neonate/Infant | Complicated | 7 | 9.951±1.12$^{abc}$ | 3.06±0.67$^{a}$ | 5.89±5.24 | 0.59±0.39 | 0.39±0.18$^{a}$ | 0.03±0.02 |
| | Uncomplicated | 133 | 9.18±0.26$^{b}$ | 3.12±0.15$^{bc}$ | 5.22±1.20 | 0.61±0.09 | 0.43±0.19$^{b}$ | 0.03±0.00 |
| | Control | 112 | 11.53±0.28$^{a}$ | 4.32±0.17$^{ab}$ | 5.92±1.31 | 0.82±0.10 | 0.37±0.78$^{c}$ | 0.04±0.00 |
| Children | Complicated | 15 | 10.28±1.72$^{abc}$ | 6.17±1.02$^{b}$ | 4.42±3.58 | 0.66±0.26 | 0.35±0.53$^{abc}$ | 0.03±0.01 |
| | Uncomplicated | 266 | 8.25±0.57$^{bc}$ | 4.28±0.34$^{ab}$ | 5.51±0.85 | 0.64±0.06 | 0.54±0.13$^{ab}$ | 0.03±0.00 |
| | Control | 233 | 6.78±0.58$^{c}$ | 3.53±0.35$^{abc}$ | 3.55±0.91 | 0.57±0.07 | 0.31±0.13$^{c}$ | 0.03±0.00 |
| Adolescent | Complicated | 3 | 8.51±0.77$^{bc}$ | 3.05±0.46$^{a}$ | 3.02±8.01 | 0.77±0.59 | 0.28±1.19$^{a}$ | 0.04±0.02 |
| | Uncomplicated | 27 | 7.99±0.18$^{c}$ | 3.05±0.1$^{a}$ | 3.03±2.67 | 0.64±0.20 | 0.30±0.40$^{ab}$ | 0.03±0.01 |
| | Control | 26 | 8.22±0.19$^{bc}$ | 3.71±0.12$^{abc}$ | 2.51±2.72 | 0.43±0.20 | 0.30±0.40$^{ab}$ | 0.04±0.01 |
| Test Statistics: *F-Ratio*, *Prob >F* | | | 7.72, <.0001* | 4.92, 0.0006* | 0.38, 0.8223 $^{ns}$ | 0.98, 0.4183 $^{ns}$ | 0.4934, 0.0065 * | 1.69, 0.1492 $^{ns}$ |

*Abbreviations: SD: Standard Deviation; WBC=White Blood Cell; NEU=Neutrophil; LYM=Lymphocytes; MON=Monocytes; EOS=Eosinophil; BAS=Basophil; Within parameters and across interactive measures, means ± SD with different superscripts (a, b, c, ab, bc, abc). Significance Level: *=p<0.05; ns=Not Significant (p>0.05)*

**Table 3.2. Influence of age category and malaria type on haemostatic parameters (PTT, PT, INR, ADAMTS13, VWF and FVIII) of malaria infected paediatric subjects**

| Interactive measures | | n | PTT (sec) | PT (sec) | INR | ADAMTS13 (ng/ml) | VWF (ng/ml) | FViii (ng/ml) |
|---|---|---|---|---|---|---|---|---|
| **Age category (AC)** | **Malaria type (MT)** | | | | | | | |
| Neonate/Infant | Complicated | 7 | 54.71±2.66 | 21.71±0.88 | 1.97±0.08 | 73.11±9.16 | 203.40±24.38$^{ab}$ | 137.46±20.27 |
| | Uncomplicated | 133 | 38.90±0.61 | 14.95±0.20 | 1.36±0.02 | 30.31±2.10 | 109.31±5.59$^{cd}$ | 49.61±4.65 |
| | Control | 112 | 32.93±0.66 | 13.46±0.22 | 1.23±0.02 | 18.82±2.29 | 87.19±6.10 $^{de}$ | 41.29±5.07 |
| Children | Complicated | 15 | 52.67±1.82 | 21.07±0.60 | 1.93±0.06 | 87.36±6.26 | 281.40±16.65$^{a}$ | 198.10±13.85 |
| | Uncomplicated | 266 | 39.03±0.43 | 15.60±0.14 | 1.41±0.01 | 26.17±1.49 | 108.76±3.95$^{cd}$ | 51.63±3.29 |
| | Control | 233 | 32.98±0.46 | 13.53±0.15 | 1.23±0.01 | 16.24±1.59 | 79.44±4.23$^{e}$ | 38.51±3.51 |
| Adolescent | Complicated | 3 | 55.67±4.06 | 21.33±1.34 | 1.94±0.12 | 94.25±13.99 | 264.27±37.24$^{a}$ | 224.40±30.96 |
| | Uncomplicated | 27 | 35.70±1.35 | 15.00±0.45 | 1.36±0.04 | 31.27±4.66 | 130.42±12.41$^{ac}$ | 60.59±10.32 |
| | Control | 26 | 32.23±0.38 | 13.77±0.46 | 1.25±0.04 | 18.34±4.75 | 78.57±12.65$^{cde}$ | 34.68±10.52 |
| Test Statistics: F-Ratio, Prob >F | | | 0.759, 0.5519 $^{ns}$ | 1.100, 0.3533 $^{ns}$ | 0.621, 0.6480 $^{ns}$ | 0.775, 0.5418 $^{ns}$ | 2.46,0.0444* | 2.197, 0.0676 $^{ns}$ |

*Abbreviations: SD: Standard Deviation; PTT: Partial Thromboplastin Time; PT: Prothrombin Time; INR: International Normalized Ratio; ADAMTS13: A Disintegrin and Metalloproteinase with a Thrombospondin Type 1 motif, member 13;VWF: Von Willebrand Factor and FVIII: Factor VIII. Within parameters and across interactive measures, means ± SD with different superscripts (a, ab, ac, cd, de, cde, e). Significance Level: *=p<0.05; ns=Not Significant (p>0.05)*

**Table 3.3. Influence of age category and malaria type on haemostatic parameters (fbg, l-arginine and d2d) of malaria infected paediatric subjects**

| Interactive measures | | N | FBG (mg/ml) | L-Arginine (pg/ml) | D2D (pg/ml) |
|---|---|---|---|---|---|
| **Age Category** | **Malaria Type** | | | | |
| Neonate/Infant | Complicated | 7 | 15.29±2.43$^{bc}$ | 56.34±12.23 | 3570.71±717.50$^{b}$ |
| | Uncomplicated | 133 | 6.99±0.56$^{d}$ | 50.76±2.81 | 2014.55±164.61$^{b}$ |
| | Control | 112 | 4.20±0.61$^{ef}$ | 41.39±3.06 | 1762.47±179.38$^{b}$ |
| Children | Complicated | 15 | 25.56±1.66$^{a}$ | 27.64±8.36 | 6929.07±490.15$^{a}$ |
| | Uncomplicated | 266 | 6.59±0.39$^{d}$ | 55.43±1.98 | 2281.37±116.39$^{b}$ |
| | Control | 233 | 3.81±0.42$^{f}$ | 48.78±2.12 | 1850.57±124.36$^{b}$ |
| Adolescent | Complicated | 3 | 28.40±3.72$^{ab}$ | 29.12±18.69 | 3980.00±1096.00$^{ab}$ |
| | Uncomplicated | 27 | 8.46±1.24$^{cde}$ | 47.58±6.23 | 3002.61±365.33$^{b}$ |
| | Control | 26 | 3.98±1.26$^{def}$ | 36.67±6.35 | 1930.78±372.29$^{b}$ |
| Test Statistics: *F-Ratio*, *Prob >F* | | | 3.815, 0.0044* | 1.432, 0.2214 $^{ns}$ | 4.707, 0.0009* |

*Abbreviations: SD: Standard Deviation; FBG: Fibrinogen; D2D: D-dimer*

*Within parameters and across interactive measures, means ± SD with different superscripts (a, d, f, ab, bc, cde, def, ef, f). Significance Level: *=p<0.05; ns=Not Significant (p>0.05)*

## 3. RESULTS AND DISCUSSION

Influence of age and malaria parasite on haematological parameter was found to have significant difference. The mean absolute count of WBC in neonate with complicated and uncomplicated malaria was observed to have a significant decrease when compared with the control subjects (Table 3.1). This is consistent with the findings of [25], but in contrast with the finding of [26], as well as a significant increase in total white blood cell (TWBC) in children and adolescent with complicated malaria in line with the findings of [27]. White blood cells (WBC) count in the body can vary during the different stages of malaria infection. Leucopenia is common during acute malaria parasite.

A significant decrease in absolute neutrophil value in neonate was observed between the complicated/uncomplicated vs control groups. This finding is found to contrast that of a previous study which reported that malaria induced changes in haematologic parameters including an increase in neutrophil count [26] but in line with the findings of [25].

This may be due to the phagocytic nature of neutrophil to combat infection and get destroyed in the process. However, a significant increase was observed in children in line with [26] indicating the role of age in disease pathogenesis. A significant decrease in absolute eosinophil in complicated malaria subjects was seen when compared to uncomplicated malaria subjects in line with the finding of [26]. This appeared that decrease in eosinophil count could be due to tissue sequestration and destruction rather than decreased production.

Age was found to have no significant influence on PTT, PT, INR, FVIII, ADAMTS13 and L-arginine (Table 3.2) in complicated and uncomplicated malaria parasite infected subjects in contrast with [27] but supported by the findings of [2]. This could be due to the compensated state influenced by age factor. Activation of the coagulation cascade as a consequence of inflammation is an essential part of the host defense of the body in an effort to contain the invading entity and to keep the consequent inflammatory response to a limited area. Trends between increase in VWF and malaria parasite type were observed to be influenced by age in this study. VWF was significantly higher in subjects with complicated malaria compared to those with uncomplicated malaria in the different age group. More so, VWF across parameters was seen to be highest in children followed by adolescent and then neonate. This finding is consisted with the notion of [28,29]. This report confirmed the view that *Plasmodium* infection pathogenesis is age dependent. A significant increase in fibrinogen was seen in line with the report of [5]. This report indicates an in vivo activation of platelet with controlled response. Likewise a significant linear increase in D2D (Table 3.3) was observed to be influenced by age in complicated and uncomplicated malaria parasite in line with the report of [30]. This report indicate that D-dimer levels are increased in the circulation of cases of *falciparum*-infected subjects in complicated and uncomplicated malaria parasite that are sensitive to fibrin formation and fibrinolysis by plasmin.

## 4. CONCLUSION

In conclusion, haematological parameters decreased as malaria got complicated in neonates, increased in children, while haemostatic parameters such as VWF, FBG and D2D increased in the three age categories being highest in children.

## CONSENT

All authors declare that written informed consent was obtained from the patient (or other approved parties) for publication of this case report and accompanying images.

## ETHICAL APPROVAL

All authors hereby declare that all experiments have been examined and approved by the appropriate ethics committee and have therefore been performed in accordance with the ethical standards laid down in the 1964 Declaration of Helsinki.

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## COMPETING INTERESTS

Authors have declared that no competing interests exist.

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