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Some Haemostatic Parameters among Sickle Cell Disease Children during Stable and Crisis States in Sokoto, North Western, NigeriaIbrahim, A.B. ^{*1}, Muhammad, H.B. ¹, Erhabor, O. ¹, Udomah, F.P. ¹, Jiya, N.M. ², Isah, D. ¹, Ibrahim, K.K. ¹, Buhari, H.A. ¹ and Wasagu, I.Z. ³

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Abstract

Sickle Cell Disease (SCD) is a group of blood genetic disorders that result in sickling of Red Blood Cells (RBC) when they are under low oxygen tension due to the mutation in the oxygen-carrying protein haemoglobin (haemoglobin S) found in red blood cells. This case control study was designed to determine the levels of some haemostatic parameters (PT and PTTK) of children with SCD (HbSS) in Sokoto, Nigeria. The subjects for this study included 72 children with SCD (36 in steady state and 36 in crisis). Thirty-six (36) age and gender-matched haemoglobin AA children were monitored as controls. Four and half (4.5ml) of blood was collected from both the SCD subjects (tests) and non-SCD (controls) by venipuncture and delivered into plastic tube containing 0.5ml trisodium citrate. The anticoagulated blood was centrifuged at 1000g for 10 minutes and the platelet poor plasma was removed and placed in plastic tube which was then used to determine the PTTK and PT manually using the Diagen PTTK and PT Kit (Diagen, Diagnostic Reagents, United Kingdom). Testing was carried out following the manufacturers' standard operating procedures and processed at the Haematology Laboratory of Usmanu Danfodiyo University Teaching Hospital, Sokoto, Nigeria. The mean age of the participants in steady, crisis and control groups are 7.08±4.4, 8.81±4.0, and 8.17±3.5 years respectively. The results showed that majority of SCD subjects were aged 5-8 years (29.2%), Hausa ethnic group (65.3%) and females (52.8%). The mean PT and PTTK of SCD subjects on steady state (18.8 ± 1.6 and 39.5 ± 1.7 seconds) and on crisis (18.6 ± 1.9 and 39.6

± 2.0 seconds) were significantly prolonged (p<0.05) compared to those of subjects with HbAA (15.5 ± 1.3 and 37.8 ± 1.5 seconds). The mean PT was slightly prolonged in steady group (18.8 ± 1.6) than in crisis (18.6 ± 1.9) while the mean PTTK was marginally prolonged in crisis (39.6 ± 2.0) than in steady group (39.5 ± 1.7 sec) when compared between the two states. However, both PT and PTTK difference were not statistically significant (p=0.61 and p=0.95). This study showed that the levels of PT and PTTK are significantly prolonged among SCD subjects when compared with control subjects.

Keywords: PT, PTTK, SCD, Steady, Crisis, Control and Sokoto.

Introduction

Sickle cell disease (SCD) is an autosomal recessive genetic disease of the red blood cells (RBCs) haemoglobin that result from homozygous or compound heterozygous inheritance of mutation in the β -globin gene (National Heart, Lung Blood Institute, 2015). Haemoglobin is the protein in red blood cells that carries oxygen and structurally it is normally made of two alpha globin chains and two beta globin chains or subunits (Stein, *et al.*, 2006). In people with anaemia, at least one of the beta-globin subunits in haemoglobin is replaced by any of these abnormal versions of beta-globin which can distort RBCs into a sickle shape (Erhabor and Adias, 2013). Sickle Cell Disease (SCD) is characterized by sickling that Red Blood Cells (RBC) undergo when they become deoxygenated as a result of an abnormality in the oxygen-carrying protein haemoglobin

(haemoglobin S) found in red blood cells (Erhabor *et al.*, 2010). This condition affects about 4.4 million people globally while an additional 43 million have sickle-cell trait (Global Burden of Disease, 2015) and is most prevalent in people from sub-Saharan Africa; South America, the Caribbean, Central America, Saudi Arabia, India and the Mediterranean (World Health Organization, 2017). It is reported that 20% of Nigerians currently have Sickle cell gene (Federal Ministry of Health Nigeria, 2018). SCD is associated with RBC having shorter lifespan (less than 100-120 days), reduced oxygen carrying capacity, vaso-occlusion, inflammation as well as impairment in iron metabolism (Serjeant, 2010). The etiology of SCD result from homozygous or compound heterozygous inheritance of mutation in the β -globin gene. The resulting substitution of the hydrophilic amino acid glutamic acid at the sixth position by the hydrophobic amino acid valine, leads to the production of haemoglobin S (HbS). HbS polymerizes when deoxygenated and this polymerization is associated with cell dehydration and increased red cell density (Junior *et al.*, 2012). This result in the pathogenic change in shape of the erythrocytes to the sickle shape, which is stiff, deforms poorly and markedly increases blood viscosity. The most common form of SCD is sickle cell anaemia in which the sickle beta globin gene is inherited in a homozygous state (HbSS). Other forms of SCD include compound heterozygous states for the sickle beta globin gene and HbC (HbSC), B-thalassemia (HbSB-thal.), HbD (HbSD), HbO (HbSO) or Hb Lepore (Hb Lepore) (Erhabor and Adias, 2013).

Clinical problems encountered in SCD relate to vaso-occlusion, caused due to the dense, rigid and sickling red cells and impaired blood flow via microvascular blockage by sickled erythrocytes that adhere to the endothelium leading to widespread microvascular damage, tissue necrosis and infarction affecting the bones and other tissues, and is thought to underlie acute (painful episodes, acute chest syndrome and chronic avascular necrosis, renal insufficiency complications of the disease (Jiya *et al.*, 2017). Also, intracellular polymerization ultimately damages the red cell membrane and leads to

chronic and episodic extra-vascular and intravascular Haemolytic anaemia. Haemolysis linked nitric oxide (NO) dysregulation and endothelial dysfunction results in leg ulcer, pulmonary hypertension (PHTN), priapism and stroke (Junior *et al.*, 2012).

Haemostasis is the combination of cellular and biochemical events that function together to keep blood in the liquid state within the veins and arteries and prevent blood loss following injury through the formation of a blood clot (Delvaeye and Conway, 2009). It consists of a complex regulated system which is dependent on a delicate balance among several systems which include the vascular system, coagulation system, fibrinolytic system, platelets, kinin system, serine protease inhibitors, and the complement system. The systems work together when the blood vessel endothelial lining is disrupted by mechanical trauma, physical agents, or chemical trauma to produce clots. The clots stop bleeding and are eventually dissolved through the fibrinolytic process. As a result, there is a delicate balance between the production and dissolution of clot during the Haemostatic process (Erhabor, *et al.*, 2014). A disruption of this balance may precipitate thrombosis or haemorrhage as a result of hypercoagulation or hypocoagulation, respectively (Verhamme and Hoylaerts, 2009).

According to Erhabor and Adias (2013), Haemostasis is categorized as either a primary or secondary process. Primary Haemostasis involves the response of the vascular system and platelets to vessel injury. It takes place when there are injuries to small vessels during which the affected vessels contract to seal off the wound and platelets are mobilized, adhere and aggregate to components of the sub endothelium of the vasculature. Platelet adhesion requires the presence of various factors such as von Willebrand factor (vWF) and platelet receptors (IIb/IIIa and Ib/IX). Additional platelets are attracted to the site of injury by the release of platelet granular contents, such as adenosine diphosphate (ADP). The platelet plug is stabilized by interaction with fibrinogen. Thus, a defect in platelet function or von Willebrand disease (vWD) may result in debilitating and

sometimes fatal haemorrhage. Secondary Haemostasis involves the response of the coagulation system to vessel injury. It is required to control bleeding from large wounds and is a continuation of the primary haemostatic mechanisms. Whereas the outcome of primary haemostasis is the formation of the platelet plug, the outcome of secondary Haemostasis is the formation of a thrombus (Isaac *et al.*, 2014). In hypercoagulability conditions of SCD patients' haemostatic parameters such as PT and APTT are extremely important determinants of morbidity and mortality. This research work provides evidence-based data on some haemostatic parameters in Sickle cell disease subjects in Sokoto.

Materials and Methods

Study Area

This cross-sectional study was carried out in the Department of Haematology and Paediatrics Department of Usmanu Danfodiyo University Teaching (UDUTH), Maryam Abacha Women and Children Hospital (MAWCH) and Noma Children Hospital (NCH) in Sokoto, Northwestern Nigeria. They serve as tertiary health centers for more than 5 million people of Sokoto State. The State is located in the extreme Northwestern part of Nigeria, near to the confluence of the Sokoto River and the Rima River. The State is in the dry Sahel, surrounded by sandy Savannah and isolated hills, with an annual average temperature of 28.3°C (82.9°F). Sokoto is, on the whole, a very hot area. However, maximum daytime temperatures are for most of the year generally under 40°C (104.0°F) and the dryness makes the heat bearable. The warmest months are February to April when day time temperature can exceed 45°C (113.0°F). The rainy season is from June to October during which shower are a daily occurrence. Sokoto city is a major commerce center in leather crafts and agricultural products. As at 2006, the state has a population of 3.6 million, but based on the population annual growth of 3%, the calculated projected population for Sokoto State is now standing at around 5.3 million (National Bureau of Statistics, 2018).

Study Subjects

The population for this study comprised of Hb Electrophoresis confirmed Children with SCD (with HbSS) who were attending the sickle cell clinic, paediatrics ward, haematology department and those presenting to the children emergency ward who fulfilled the inclusion criteria were consecutively recruited into the study. For the control group, the same method was used to recruit apparently healthy children (with HbAA) coming for follow up. The recruitment was done from UDUTH, MAWCH, and NCH in Sokoto, Nigeria.

Inclusion Criteria

The inclusion criteria for this study include; Paediatrics (from age of six months to sixteen years) Hb Electrophoresis confirmed HbSS and parent/guardian willingness to sign a written informed consent for their wards to participate in the study as subjects.

Exclusion Criteria

The following were excluded from participating in this study; sickle cell subjects < 6 months or > 16 years, individuals who are not known to have SCD and children whose parents/guardians refused to sign a written informed consent for their wards to participate in the study as subjects were excluded from the study.

Sample Size Determination

Sample size of the study was determined using the standard formula for the calculation of minimum sample size:

$$n = Z^2 Pq / d^2$$

Where:

- n = minimum sample size
- z = standard normal deviation and probability.
- p = prevalence or proportion of value to be estimated from previous studies.
- q = Proportion of failure (= 1 - P)
- d = precision, tolerance limit, the minimum is 0.05.

$$\text{Therefore } n = z^2 pq / d^2$$

$$\text{Where } Z = 95\% (1.96)$$

$$P = 5\% (0.05) \text{ is the prevalence HbSS in Sokoto (Jiya } et al., 2017).$$

$$q = 1 - 0.05 (= 0.95)$$

$$d = 5\% (0.05)$$

Therefore $n = (1.96)^2 (0.05)(0.95) / (0.05)^2$
 $n = 72$

Study Design

This is a case-control study designed to determine the levels of some haemostatic parameters among SCD subjects in Sokoto. A total of one hundred and eight (108) paediatric subjects were recruited for this study, in which clinically confirmed seventy-two (72) SCD (with HbSS) subjects including 36 on steady state and 36 on crisis. Thirty-six (36) were non-SCD (HbAA) apparently healthy coming for follow up will be serving as the control group.

Ethical Clearance

Ethical approval was obtained from the Hospitals Ethical Committees and informed consent was obtained from all the subjects or their guardians before sample collection.

Questionnaire

A semi structured questionnaire was administered to all consenting participants to obtain information on subjects' bio-data and socio-demographic characteristics.

Informed Consent

Written informed consent was obtained from all the eligible participants in respect to their approval.

Sample Collection

Four and half milliliters (4.5ml) of blood was collected from both the SCD subjects (tests) and non-SCD volunteers (controls) by venipuncture and delivered into plastic tube containing 0.5ml trisodium citrate (16g/l) under strict aseptic techniques. The anticoagulated blood was centrifuged at 1000g for 10 minutes and the platelet poor plasma was removed and placed in plastic tube which was then used to determine the PTTK and PT manually using the Diagen PTTK and PT Kit (Diagen Diagnostic Reagent, UK). Testing was carried out following the manufacturers' standard operating procedures and processed at the Haematology Laboratory of School of Medical Laboratory Sciences, Usmanu Danfodiyo University Teaching Hospital (UDUTH), Sokoto, Nigeria.

Laboratory Methods

Principle of Prothrombin Time (PT) Test

The PT measures the clotting time of recalcified plasma in the presence of an optimal concentration of tissue extract (thromboplastin) and indicates the overall efficiency of the extrinsic clotting system. Although, originally thought to measure prothrombin, the test is now known to depend also on reactions with factors V, VII and X and on the fibrinogen concentration of the plasma (Diagen Diagnostic Reagent, UK).

Procedure: The reagent manufacturers procedure was followed as outlined below:

- The reagent was swirled before use
- Two hundred microlitres (200 μ L) of Calcium Rabbit Brain Thromboplastin was placed in a clotting tube within a water bath at 37°C for 2 minutes.
- One hundred microlitres (100 μ L) of citrated plasma (control) was added and a stopwatch started.
- The content of the tube was immediately tilted at regular intervals (returning to the water bath between tilting at 37°C).
- The time taken for a gel clot to form was recorded in seconds.
- The test was carried out in duplicate for both the controls and patients and the mean value for each was obtained.

Calculation: International normalized ration (INR) = {PT of test/PT of control}^{ISI}. Where ISI (International Sensitivity Index) = 1.28.

Reference range: 12 - 17 seconds

Activated Partial Thromboplastin Time (APTT) Test

Principle of the Test

The test measures the clotting time of plasma after the activation of contact factors and the addition of phospholipid and calcium chloride but without adding the tissue thromboplastin and so, indicates the overall efficiency of intrinsic pathway. To standardized the activation of contact factors, the plasma is first incubated for a set period with a contact activator such as kaolin, silica or ellargic acid. During this phase of the test, factor XIIa is produced, which cleaves factor XI to factor XIa, but coagulation does not

proceed beyond this in the absence of calcium. After recalcification, factor XIa activates factor IX and coagulation follows. A standardized phospholipid is provided to allow the test to be performed on PPP. The test not only depends on contact factors VIII and IX but also on the reactions with factors X, V, prothrombin and fibrinogen. It is also sensitive to the presence of circulating anticoagulants (inhibitors) and heparin (Diagen Diagnostic Reagent, UK).

Procedure: The reagent manufacturers procedure was followed as outlined below:

- The reagent was swirled before use.
- Two hundred microlitres (200 μ L) of Kaolin Platelet Substitute Mixture was pipetted into a cleaned glass test tube and incubated in water bath at 37°C for 2 minutes.
- One hundred microlitres (100 μ L) of citrated plasma (control) was added into the glass tube and was tilted at interval for 2 minutes.
- One hundred microlitres (100 μ L) of 0.025 M of calcium chloride (pre-incubated at 37°C) was added and a stopwatch started.
- The content of the tube was immediately tilted at regular intervals (returning to the water bath between tilting).
- The time taken for a gel clot to form was recorded in seconds.
- The test was carried out in duplicate for both the controls and patients and the mean value for each was obtained.
- The test was carried out in duplicate on the patient's plasma and the control plasma.

Reference range: 35-43 seconds (Diagen Diagnostic Reagent, UK).

Statistical Analysis

Data collected was recorded on an excel spreadsheet and later subjected to statistical analysis using a statistical software SPSS version 23. Chi-square descriptive analysis was used to define the nature of the distribution of data for the test and control groups based on sociodemographic factors. The independent t-test and paired t-test and ANOVA were used to determine significant difference of PT and PTTK levels between test and control subjects, also in

steady state and crisis among SCD subjects. The results were expressed as mean \pm SD, percentage and presented in tables and Statistical significance level was considered at $p \leq 0.05$.

Results

The results of the analysis of sociodemographic factors of 108 participants and some haemostatic parameters (PT and PTTK) of 72 SCD subjects (36 on crisis and 36 on steady state) and 36 HBAA controls. Table 1 shows the socio-demographic characteristic of study subjects and control subjects. The mean \pm SD of age participants on crisis, steady and control groups are 7.08 \pm 4.4, 8.81 \pm 4.0 and 8.17 \pm 3.5 years respectively. The majority of the study subjects were aged 5-8 years 33 (30.6%), of Hausa ethnic group 66 (61.1%) and females 57 (52.8%). Table 2 shows the comparison of the Mean \pm SD of PT and PTTK levels of SCD subjects in steady, crisis state and control groups. The mean \pm SD of PT and PTTK of subjects with SCA in steady state as well as those during crisis were prolonged compared to the HBAA controls. There was a statistically significant difference in the mean values of both PT and PTTK between SCA subjects and the controls ($p=0.000$ and $p=0.004$ respectively). Table 3 shows Comparison of the Mean \pm SD of PT and PTTK levels of study subjects in steady and crisis state. The mean PT was slightly prolonged in steady group (18.8 \pm 1.6) than in VOC (18.6 \pm 1.9) while the mean PTTK was marginally prolonged in VOC group (39.6 \pm 2.0) than in steady group (39.5 \pm 1.7 sec). However, both PT and PTTK difference were statistically not significant ($p=0.61$ and $p=0.95$). Table 4 shows the interaction between sociodemographic factors and levels of PT and PTTK of SCD subjects. There was a no significant difference in the mean PTTK levels of ($p=0.481$) and PT ($p=0.316$) based on age groups. The PT levels in males was higher than in females SCD patients, but there was no statistically significant difference ($p=0.173$). There was a significant difference ($p=0.018$) in the levels of PTTK based on gender. There was no statistically significant difference in the levels of PT ($p=0.316$) and PTTK ($p=0.284$) based on ethnicity.

Table 1: Socio-Demographic Characteristic of Study Subjects and Control subjects

Age Range (Years)	Steady N=36(%)	VOC N=36(%)	Control N=36(%)	Total N=108(%)	p-value
≤ 4	12(33.3%)	6(16.7%)	8(22.2%)	26(24.0%)	0.62
5 – 8	11(30.6%)	10(27.8%)	12(33.3%)	33(30.6%)	
9 – 12	7(19.4%)	12(33.3%)	11(30.6%)	30(27.8%)	
13 – 16	6(16.7%)	8(22.2%)	5(13.9%)	19(17.6%)	
Ethnicity					
Hausa	23(63.9%)	24(66.7%)	19(52.8%)	66(61.1%)	0.22
Fulani	6(16.7%)	5(13.9%)	11(30.6%)	22(20.4%)	
Yoruba	1(2.8%)	3(8.3%)	4(11.1%)	8 (7.4%)	
Igbo	2(5.5%)	3(8.3%)	2(5.5%)	7 (6.5%)	
Others	4(11.1%)	1(2.8%)	0(0.0%)	5 (4.6%)	
Gender					
Male	16(44%)	18(50%)	17(47%)	51(47.2)	0.89
Female	20(56%)	18(50%)	19 (53%)	57(52.8)	

Key: PT = Prothrombin Time, PTTK = Partial Thromboplastin Time with Kaolin, p = Statistically Significant ($p > 0.05$)

Table 2: Comparison of Mean±SD of PT and PTTK levels of Study subject in Steady, Crisis state and Control groups

	Steady	VOC	Control	p-value
PT (seconds)	18.8 ± 1.6	18.6 ± 1.9	15.5 ± 1.3	0.000
PTTK (seconds)	39.5 ± 1.7	39.6 ± 2.0	37.8 ± 1.5	0.004

Key: VOC= vaso-occlusive crisis, PT = Prothrombin Time, PTT = Partial Thromboplastin Time with Kaolin, p = Statistically Significant ($p < 0.05$)

Table 3: Comparison of Mean±SD of PT and PTTK levels of Study subjects in Steady and Crisis state.

	Steady	VOC	P-value
PT (seconds)	18.8 ± 1.6	18.6 ± 1.9	0.61
PTTK (seconds)	39.5 ± 1.7	39.6 ± 2.0	0.95

Key: PT = Prothrombin Time, PTT = Partial Thromboplastin Time with Kaolin, p = Statistically not Significant ($p > 0.05$).

Table 4: Interaction between Sociodemographic factors and levels of PT and PTTK of SCD subjects.

Sociodemographic Factors		PT Mean ± SD	PTTK Mean ± SD
Age Range	0 – 4	18.28 ± 1.5	39.33 ± 1.2
	5 – 8	18.43 ± 2.2	39.44 ± 2.2
	9 – 12	19.31 ± 1.4	39.84 ± 1.6
	13 – 16	19.07 ± 1.9	40.00 ± 2.4
	p-value	0.316	0.481
	χ^2	1.498	2.671
Gender	Male (34)	19.03 ± 1.8	40.09 ± 1.6
	Female (38)	18.45 ± 1.8	39.05 ± 2.0
	p-value	0.173	0.018
	χ^2	3.758	2.464
Ethnicity	Hausa (47)	18.66 ± 1.9	39.34 ± 2.0
	Fulani (11)	18.82 ± 1.5	39.82 ± 1.5
	Yoruba (4)	17.75 ± 1.0	39.00 ± 1.4
	Igbo (5)	20.20 ± 2.4	41.20 ± 1.8
	Others (5)	18.40 ± 1.5	39.60 ± 1.2
	p-value	0.316	0.284
	χ^2	5.283	3.210

Key: PT = Prothrombin Time, PTTK = Partial Thromboplastin Time with Kaolin, χ^2 = Chi-square value, p = Statistically Significant ($p < 0.05$).

Discussion

This study determined socio-demographic distribution and some haemostatic parameters among children with SCA. The sociodemographic distribution shows that the majority of SCD subjects were aged 5-8 years (29

.2%). This is in disagreement with the previous report by Erhabor *et al.* (2016) which shows that the majority SCD (35.6%) were aged below 5 years old and Jiya *et al.* (2017) also indicated that the younger children (6 months - 5.0 years) constitutes the majority of SCD patients presenting to their hospital. It also contradicts previous reports that found higher age groups for majority of SCD subjects, such as between 25 and 34 years old McClish *et al.* (2005), 25 to 34 years Levenson *et al.* (2008), 30 and 39 years Amaral *et al.* (2015) and the mean age 30 years

old (Asnani *et al.*, 2010). The highest prevalence of children with SCD in this research may be due to lack of proper genetic counseling on intending couples on the danger of sickle cell disease and the low level of awareness among the parents about sickle cell disease. The result also showed that females (52.8%) have the highest prevalence of SCA. This is in agreement with previous findings which all found that majority of SCD were females than males (Amaral *et al.*, 2015; Asnani *et al.*, 2010; McClish *et al.*, 2005; Levenson *et al.*, 2008). The previous research in Sokoto by Erhabor *et al.* (2016) and Jiya *et al.* (2017) also both indicated that the females were more affected with SCA than male children but there was no statistically significance difference. This higher prevalence rate of SCD in females in this study may be due natural distribution of females than males in the region. The ethnic

distribution among this study subjects shows that Hausa has the highest prevalence (65.3%) of SCD than other ethnic groups. This is supported by most previous researches conducted in the same region which also reported highest prevalence in Hausa. This may be due to the fact that majority of the residents in the state are Hausa speaking indigenes.

The PT which measures the performance of extrinsic pathway in conjunction with the PTTK that measures both the intrinsic and the common coagulation investigations detecting abnormalities in blood clotting (Lowe *et al.*, 2000) and can be used to monitor the treatment effects in patient at risk of thrombosis on heparin therapy (Joshua and Charles, 2008). As such shortening of the PT and PTTK predisposes patients to an increased risk of thrombosis (Korte *et al.*, 2000). This study shows a prolonged mean PT and PTTK levels in children with SCD in steady state when compared with children with normal HbAA genotype and these are further prolonged during crises. This is similar with the findings of Chinawa and Colleagues (2013) who reported prolonged mean PT and PTTK in subjects with SCA on steady state and on crisis when compared with normal HbAA individuals. Another previous work by Ataga and Orringer (2003) and Richardson *et al.* (2008) also reported prolonged PT and PTTK in SCD than in controls, which is in tandem with our findings. Furthermore, the PTTK values of all patients in steady state and those in crisis were within the reference range. However, the mean PT was not. On the other hand, the significantly prolonged PTTK of subjects in crises when compared to steady state may be due to the production of cytokines and anti-lupus factor which are elaborated during infections which in turn activate the intrinsic factors leading to their consumption (Joseph *et al.*, 2008). This in contrast to a previous finding reported by Ahmed *et al.* (2002), where the PT and PTTK values were normal among all patients in steady state, in crisis as well as among the control subjects. However, Babiker *et al.* (1987) also noted no difference between these coagulation variables among subjects and control initially, but later found prolonged levels among children with crises after the 4th day of crises. Thus, due to the

changes of coagulation levels noted, rather than being primarily responsible for the onset of crises, can only be secondary changes arising from the etiological factors of crises, i.e. stasis and acute phase proteins. Though the subject's age factor may play role in significant prolongation of the PT and PTTK observed in this work that recruited paediatric subjects (6 months to 16 years). Our observation is consistent with previous report by Chinawa *et al.* (2013), whereas Ahmed *et al.* (2002) recruited subjects between 14 - 31 years.

The findings of this study showed that there is statistical significance difference in mean values of both PT and PTTK between SCD subjects and the controls ($p < 0.05$). This is in agreement with the reports (Chinawa *et al.*, 2013; Richardson *et al.*, 2008; Ataga and Orringer, 2003; Babiker *et al.*, 1987). This may be due to high prolonged variation in mean PT and PTTK levels in SCD subjects than the controls in these researches. Thus, our finding is in contrast with the report by Ahmed *et al.* (2002) who found no significant difference in the levels PT and PTTK among SCD subjects and controls.

The mean PT level was marginally higher in age group of 9 - 12 years and also the mean PTTK level was higher in 13 - 16 years. There was a no significant difference in the mean PTTK levels of ($p > 0.05$) based on age groups. This is in disagreement with the report by Chinawa *et al.* (2013) which indicated that PT and PTTK values were higher in 3-5 years group and lowest in the 12-14 age groups, this differences in the PT and PTTK did not change significantly based on age. These findings are consistent with previous reports of Joseph *et al.* (2008) who observed that the mean values of the haemostatic parameters did not vary significantly based on age. This may be attributed to the liver that is responsible for the production of most of the clotting factors is impaired among the age groups. The PT levels between male and female SCD patients was prolonged but there was no significant difference ($p > 0.05$). However, there was a significant difference ($p < 0.05$) in the levels of PTTK of Males and Females. There was no significant difference in the levels of PT and PTTK ($p > 0.05$) based on ethnicity, though PT and PTTK were higher in Igbo ethnic groups.

Conclusion

The study showed that children with sickle cell anaemia have significantly prolonged PT and PTTK compared to those with normal haemoglobin AA. The mean PT was slightly prolonged in steady state group than in crisis while the mean PTTK was a little bit prolonged in crisis than in steady group. Majority of the study subjects in this study are females.

Recommendations

Based on the results observed in this study, we recommend that children with SCD be screened for these coagulation parameters especially when in vaso-occlusive crises. Intending couples should be screened for SCD and counselled on the danger of this disease.

Conflict of Interest: The authors have declared that there is no conflict of interest associated with this study.

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