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SICKLE OR NO SICKLE: THE LIFE OF THE FLESH IS IN THE BLOOD; NO BLOOD NO LIFE

By

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INTRODUCTION

Mr. Vice-Chancellor, Sir, it is with great joy that I stand before this audience to give the first inaugural lecture for this year 2019, the Inaugural Lecture Series 330. Happy New Year. I give God Almighty all the glory and honour for keeping us alive to see this day. Today I stand here to present the second inaugural lecture from the Department of Haematology and Immunology, which I dedicate to my late parents Gladys and Joseph, who invested in my education; my little sister, Omolara, who died at the age of 12 years from complications of sickle cell disease (SCD); and all individuals living with this disease in Nigeria.

I am the first born of the Bamgbaiye family that hails from Okero, Otan Aiyegbaju, Osun Northeast, Osun State. My mother had seven children with one stillbirth and four alive today. On hind sight it is possible that the children that died before her fifth birthday had sickle cell disease. In 1963 my junior brother, and I were taken to London, UK, to join our parents who had previously left Nigeria in search of the Golden Fleece. In 1974 I returned with some members of my family to Nigeria with four 'A' Levels at the instruction of my father who declined my admission into The City University, London, to study Chemistry because he wanted me to study medicine at the University of Ife (now Obafemi Awolowo University) and not the UK.

I was interviewed by the late Prof. TAI Grillo (of blessed memory) and admitted into direct preliminary year of Medicine because he said that I needed time to acclimatize. The admission process was different in those days, but thank God I graduated as a medical doctor in 1981 and went onto residency training in Internal Medicine (graduated 1994) and later Pathology (Haematology; 1998) at Obafemi Awolowo University Teaching Hospitals Complex (OAUTHC), Ile-Ife. As a senior registrar in Internal Medicine I took time out to subspecialise in Haematology at the Hammersmith Hospital (Dip. Haem.; Imperial College, University of London) in 1989. This programme was converted to an MSc degree programme through the intervention of my set, the 1989 set, through a petition to the Senate of the University of London. Then God's favour took me to the University of Birmingham where I obtained a PhD in July 1992 having studied the 'Rheology of Vaso-occlusion in Sickle Cell Disease', as a Wellcome Trust Scholar. I returned to Nigeria immediately and was employed as a Lecturer 1 in the Department of Haematology and Immunology, Faculty of Basic Medical Sciences, by Prof. Vincent Nwuga (also of blessed memory) who was the Dean at the time. My vision was to

establish a haematopoietic cell transplantation unit at OAU teaching hospital, Ile-Ife, thereby providing a cure for many haematological diseases.

This inaugural lecture is a compilation of my contributions to the knowledge and practice of my calling (my profession) in Haematology and Blood Transfusion, through teaching, research and service; and to present my vision for future research outputs to improving the quality of care and quality of life of my patients, particularly those with sickle cell disease. The title of this lecture is,

"Sickle or No Sickle: the Life of the Flesh is in the Blood; No Blood No Life".

Mr. Vice-Chancellor Sir, to begin with, it has been written in Leviticus 17: 14b that, "for the life of all flesh is the blood thereof" (KJV), therefore, blood is life.

WHAT IS HAEMATOLOGY?

Haematology is the study of blood and blood disorders/diseases. The blood is arguably the most important tissue in the body because it is responsible for the delivery of oxygen from the lungs and nutrients from the gut to other parts of the body as long as the heart is prepared to pump it. The blood is also responsible for transporting waste products of cell metabolism to their excretory sites e.g. lungs, gut, kidneys, liver, skin etc. appropriately.

THE BLOOD

The blood is circulated around the body in vessels called arteries and returned to the lungs in veins (Figure 1). Arterial blood is bright red because it is rich in oxygen (O_2), while venous blood is dark red because the oxygen level is low and is high in carbon dioxide (CO_2).

Blood contains formed elements (cells) and plasma (liquid component; Figure 2). The cells in blood are red cells, white cells and platelets (Figure 3) and they will form the basis of this lecture.

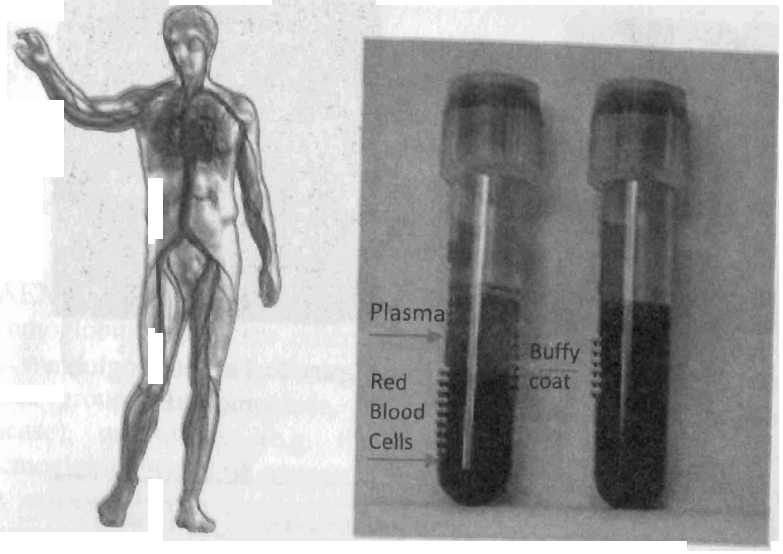

Figure 1: Arteries and Veins **Figure 2:** Components of blood

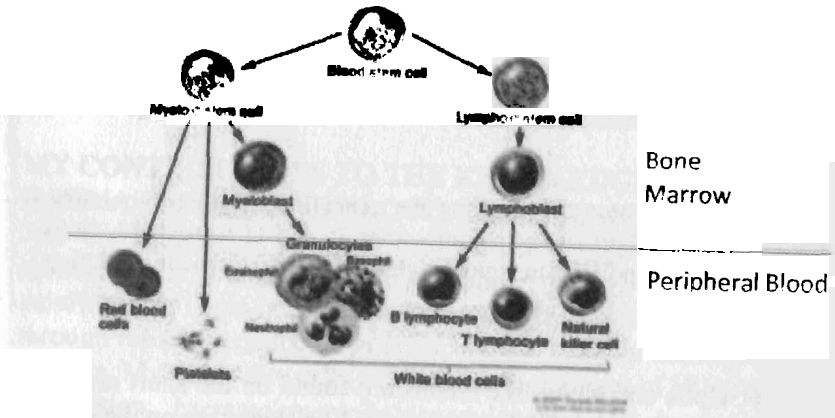

Figure 3: The formed elements of blood (blood cells)

Menghini in 1747 discovered that blood contained iron by showing that iron-like particles could be extracted with the use of a magnet from burnt blood. The red cells are red (Figure 4) because they contain haemoglobin (Hb), a protein molecule with a tetrameric structure composed of two alpha (α) or alpha-like (ζ) polypeptide chains and two non-α chains that may be β, δ, γ, ε and ζ folded around a haem moiety (Figure 5) depending on the gestational age.

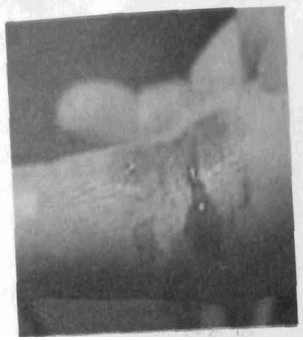

Figure 4: The life of the flesh is in the blood

Figure 5: The Haemoglobin molecule

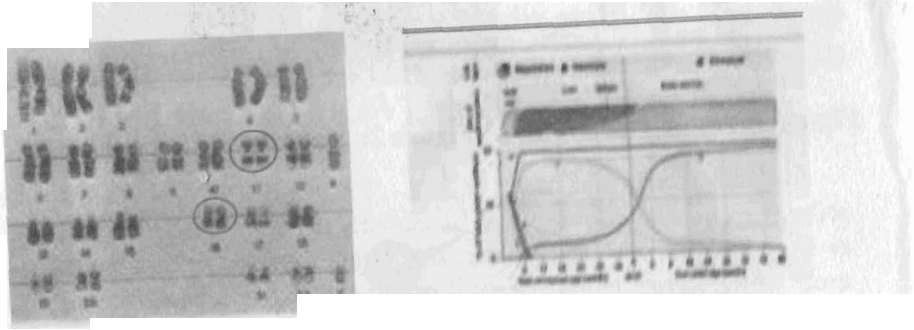

Figure 6: Human chromosomes showing the genes for alpha and non-alpha chains are found on the short arm of chromosomes 16 and 11 respectively (left) and the site of haematopoiesis and globin gene expression switching during development (right).

The genes for alpha and alpha-like chains are found on the short arm of chromosome 16, while the genes for non-alpha-like chains are on the short arm of chromosome 11 (Figure 6). The usual (major) adult haemoglobin A is composed of $\alpha_2\beta_2$, foetal heamoglobin (Hb F) is composed of $\alpha_2\gamma_2$ and the minor adult haemoglobin (Hb A_2) is composed of $\alpha_2\delta_2$.

Haemoglobin phenotype is the expression of the genes (genotype) that code for haemoglobin (Hb). The production of haemoglobin commences in the yoke sac in the first few weeks of life, then in the liver, then the spleen and finally in the bone marrow. The production of foetal haemoglobin is switched off at birth and the major adult Hb increases and reaches adult levels by the age of six months (Figure 6).

HAEMOGLOBINOPATHY

Haemoglobinopathies are inherited disorders of haemoglobin synthesis that may or may not have clinical significance. They are divided into five broad groups which include, structural (qualitative; e.g. sickle cell disease), quantitative (e.g. thalassaemias), unstable haemoglobins, haemoglobins with low oxygen affinity and the M-haemoglobins which are associated with familial cyanosis, while haemoglobins with high oxygen affinity cause increased production of red cells (polycythaemia). Haemoglobinopathies that are not associated with high oxygen affinity cause shortage of blood (anaemia), consequent upon the reduction of red cell live span from the usual 120 days to 5-25 days. These haemoglobinopathies are characterized by an uncompensated anaemia with unconjugated hyperbilirubinaemia (lemon yellow jaundice) and reticulocytosis.

MY CONTRIBUTIONS TO THE KNOWLEDGE OF HAEMATOLOGY AND BLOOD TRANSFUSION

Mr. Vice-Chancellor Sir, my major thrust in research is in sickle cell disease where I contributed to the understanding of the steady state, painful crisis (vaso-occlusion) and certain organ complications in SCD through research conducted in collaboration with my colleagues on adult patients. My minor thrusts in research are in the areas of HIV/AIDS, blood transfusion, haemato-oncology, coagulopathy and haemopoietic cell transplantation.

SICKLE CELL DISORDER

Sickle cell disorder is the inheritance of a single sickle haemoglobin gene or more and it includes Hb AS, while sickle cell disease is the inheritance of a sickle haemoglobin gene together with another unusual haemoglobin gene (variant), which may be in the homozygous state (Hb SS; sickle cell anaemia; SCA) or a compound state (Table 1). Sickle cell anaemia was first reported in 1910 by James Herrick in Chicago, USA, but the term

"sickle cell anaemia" was first used by Mason (1922) when describing the fourth case. Sickle cell disease has been in existence in Africa for at least five thousand years, therefore it is an ancient disease known by many names in various languages, such as, "abiku", "ogbanje", in Yoruba and "chwechweechwe" in a Ghanian language. This name calling or labelling is often associated with the myths and beliefs surrounding the disease in these areas. It was thought that affected children were reincarnated, possessed evil spirits, did not live beyond 21 years of age and could not have children of their own. All these beliefs are false because although individuals with SCD have delayed puberty, they are fertile with some even have twins and the oldest documented patient is in her nineties (Battabox.com 2016; permission from Prof. Kehinde, LUTH, Lagos, retired).

Herrick (1910) reported an anaemia characterized by bizarre, sickle-shaped cells that were first observed by a medical resident who likened the shape of the abnormal cells he saw under the microscope to a sickle. It was in the 1920's that the role of deoxygenation was discovered by Hahn and Gillespie (1927). The hereditary nature of the disease was suspected but not demonstrated until 1949 by Dr. James V. Neel. The association with haemoglobin was discovered by Linus Pauling and Harvey Itano in 1951, but the actual amino acid substitution was published by Vernon Ingram in

Table 1: Haemoglobin profile of adults with sickle cell disorder

Hb Genotype	%Hb S	%Hb A	%Hb F	%Hb A_2	Other
SS	80-95	-	2-20	N	-
SS – alpha thal	80-90	-	2-20	3.3-3.8	-
SC	40-50	-	1-4	-	C: 40-50
$S\beta^0$ thal	75-90	-	2-20	4-6	-
$S\beta^+$ thal	50-85	5-30	2-20	4-6	-
SD Punjab	40	-	2.5-5	2-3	D: 50
SO Arab	45	-	4-7	-	O Arab: 45
S Lepore	75	-	3.5-40	2	Lepore: 10
SE	60	-	4	-	E: 30-35
S/HPFH	60-70	-	25-35	1.5-2.5	-
AS (asymptomatic)	30-45	50-65	2-5	N	-

1957, thus making SCD the first disease known to man to be associated with inheritance of genes. The commonest haemoglobinopathy in man is sickle cell disease (SCD), which has a prevalence of 2-3% of the Nigerian population (Akinyanju 1989). Nigeria being the most populous black nation, has the highest burden of sickle cell anaemia (SCA) in the world (2.8 million). It is estimated that about 150,000 - 200,000 infants with SCD are born in this country every year and half would have died by the age five years and 90% before attaining adulthood if poorly managed in childhood. In addition, up to 8% are at risk of developing stroke (Verduzco, 2009).

Approximately 24% of Nigerians have the sickle cell trait (Hb AS; SCT) and can pass the gene onto their off-springs (Figure 7). Haemoglobin C, alpha- and beta-thalassaemias and other haemoglobinopathies are less common among Nigerians.

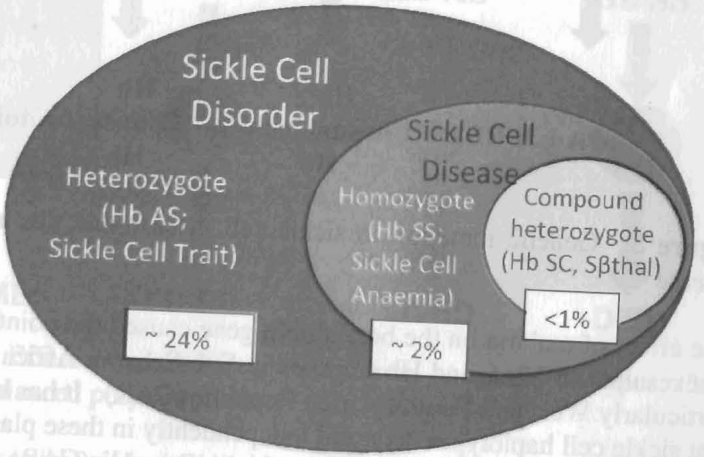

Figure 7: Schematic representation of sickle cell disorder (not to scale) showing the prevalence in Nigeria

Genetic Basis of Sickle Cell Disorder

The discovery of the sickling phenomenon in vitro by Emmel (1917) in the members of a family first suggested the genetic basis for SCD and the possibility of it being an inheritable disease. Later Huck (1923), with the detailed analysis of the pedigrees of his patients, concluded that the sickle cell phenomenon is an autosomal recessive Mendelian characteristic. Haemoglobin S (Hb S) gene is a single point mutation at position 6 from the end terminus of the beta-globin gene where amino acid glutamic acid

(GAG) is substituted by valine (GTG; Figure 8). Whereas haemoglobin C (Hb C) is caused by the substitution of lysine (AAG) for glutamic acid at the same point on the beta-globin gene. Many hundreds of thousands of years ago before the sickle cell gene mutation occurred, malaria infection was a major cause of death for people with the usual adult haemoglobin (Hb AA) living in endemic areas, because it destroyed red blood cells and caused severe anaemia.

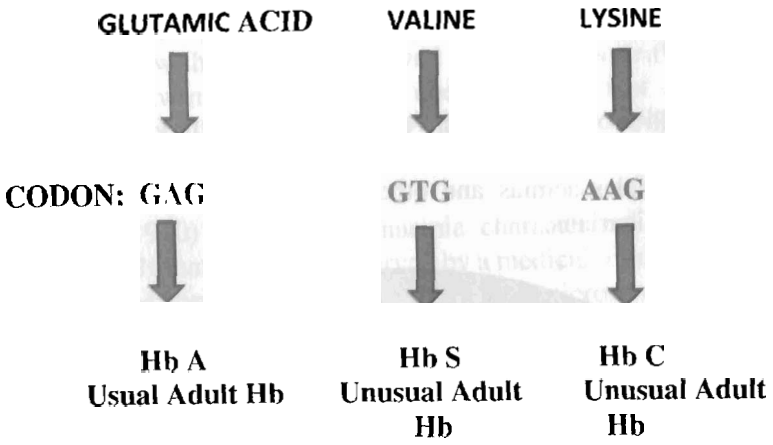

Figure 8: Genetic mutation in sickle cell disease and haemoglobin C disease

The effect of malaria on the beta globin gene caused the point mutations that resulted in Hb S and Hb C genes in Sub-Saharan Africa (Figure 9) particularly West and Central Africa Republic (CAR). It has been shown that sickle cell haplotypes occurred independently in these places and are so named; Benin, Senegal, Central African Republic(CAR), Bantu and Asian (Arabia; India). Individuals with the sickle cell trait (SCT; Hb AS) had a selective advantage and survived acute malaria (concept of balanced polymorphism (Kurnit, 1979; Soloman and Bodmer; 1979). Individuals with SCT are therefore protected from malaria infection (Figure 10) since exposure to malaria parasite causes their red cells to sickle and be removed from circulation by the reticuloendothelial system before the parasite density is high enough to cause an acute infection (Friedman 1978; Pasvol *et al* 1979). Unfortunately this is not the case for individuals with SCD in whom malaria infection worsens the anaemia and they may die (Figure 10). It has been forecasted that the sickle cell gene will persist for thousands of years even after the eradication of malaria.

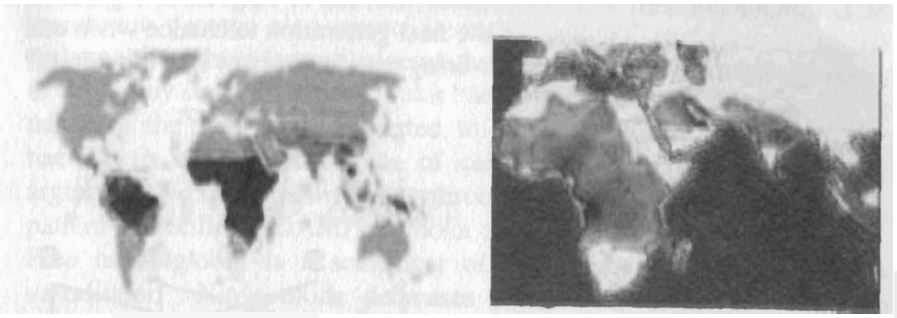

Figure 9: Modern distribution of malaria (left) and distribution of the sickle cell trait shown in pink and purple (right).

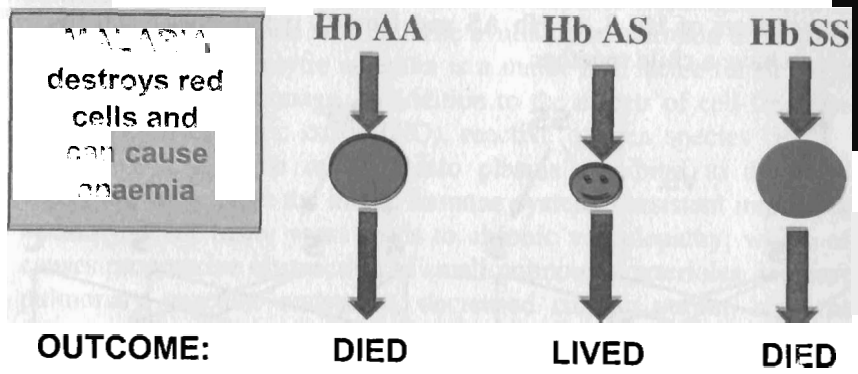

Figure 10: Effect of malaria on the common haemoglobin types, the concept of balanced polymorphism

Sickle cell disorder is inherited like the blood group, the colour of the skin, hair and eyes etc. These genes are transferred to the next generation by Mendelian fashion and for SCD, this means that if both parents have Hb AS (sickle cell trait; SCT) there is an equal chance of transferring these genes (A or S) to the offspring by each parent. Therefore, both parents are equally responsible for the birth of a child with SCA. The probability of having an affected child is one chance in four for every pregnancy (Figure 11). It is therefore possible for a family with parents at risk (i.e. having Hb AS) to have no child affected or have all or some of their children affected by the disease. Figure 12 shows that there is a 50% chance of having an affected child if an individual with SCD decides to have children with an individual with SCT. It is therefore advisable for individuals with

SCD to have children with individuals with Hb A only. It is a game of chance, so why leave the lives of the next generation to chance when one can make informed decisions not to bring pain and sorrow into the family?

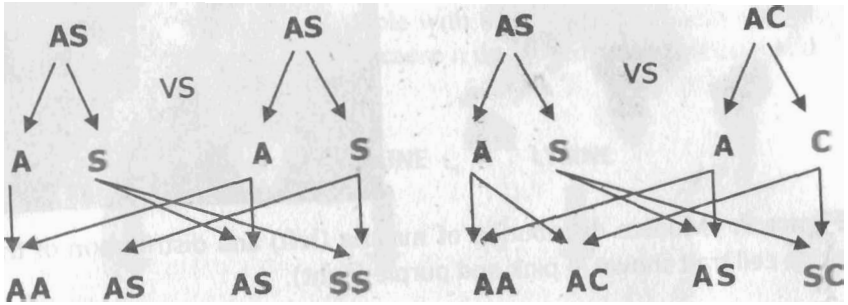

Figure 11: The probability of having an affected offspring is **one in four** if two carriers of Hb S, or Hb AS and Hb AC individuals respectively desire to have a child together.

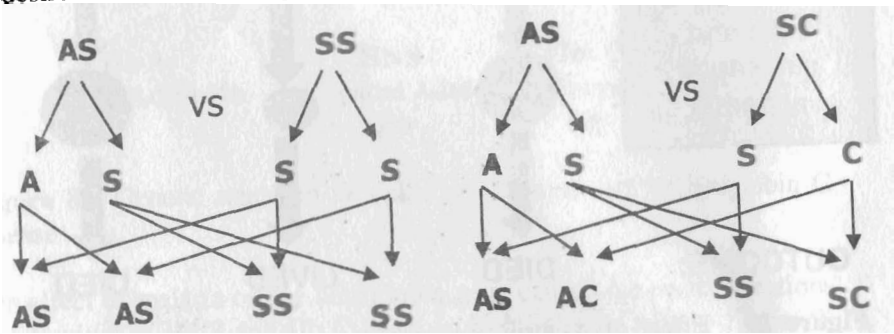

Figure 12: The probability of having an affected offspring is **one in two (50%)** if an individual with sickle cell disease (in red) desires to have a child with a carrier of the Hb S gene (Hb AS).

Sickle cell disorder is preventable

Do you know your haemoglobin type?

"The Life of the Flesh is in the Blood; No Blood No Life"

Haemolysis (Red Cell Destruction) in Sickle Cell Disease

Sickle cell disease is a chronic haemolytic disorder that may be associated with intravascular and extravascular destruction of red cells (haemolysis). When antibody-antigen reaction activates complement and sickling causes

membrane fragmentation there is intravascular haemolysis, however membrane damage also results in extravascular haemolysis of poorly deformable cells in the reticuloendothelial system by macrophages, even in the steady state. In extravascular haemolysis causes haemoglobin to be taken to the liver and converted to bilirubin, whereas, intravascular haemolysis causes the release of cell-free haemoglobin, haeme, and arginase-1, recently known as, erythrocyte damage-associated molecular pattern molecules (eDAMPs; Potoka and Gladwin, 2015), into plasma. Free haemoglobin is a scavenger of nitric oxide which is a potent vasodilator. Nitric oxide decreases platelet activation and adhesion receptor expression on vascular endothelium; decreases vascular smooth muscle proliferation; limits ischaemia-perfusion injury; modulates endothelial proliferation; and regulates inflammation (Figure 13). Therefore the higher the level of Hb F in the red cells the less severe the chronic haemolytic process and the lower the free-haemoglobin level in plasma and nitric oxide will be more available to maintain a steady state free of pain. Haemolytic anaemia is a major risk factor for developing chronic end organ damage. In addition to the effects of cell-free plasma haemoglobin on nitric oxide (NO), reactive oxygen species (ROS) are generated and haeme released into plasma can bind to the toll-like receptor-4 to activate the innate immune system. Persistent intravascular haemolysis for many years leads to chronic vasculopathy, which often causes progressive obstruction of small pulmonary arterioles, increase in pulmonary vascular resistance, decreased cardiac output, right heart failure and eventually death.

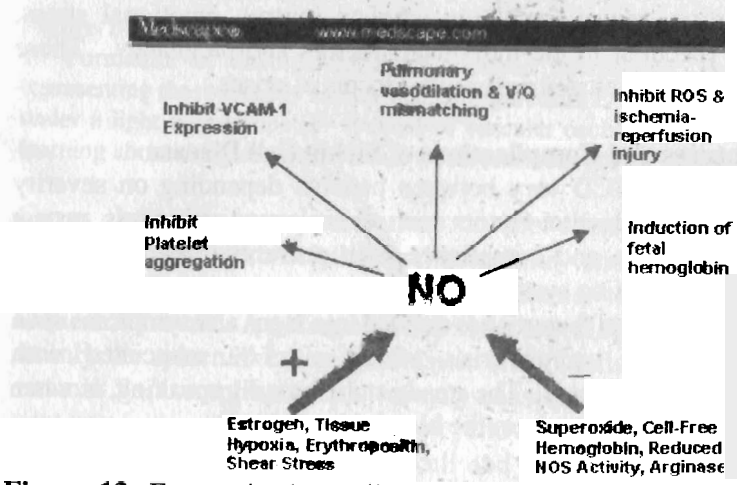

Figure 13: Factors in the inhibition and release of nitric oxide radical (Gladwin *et al*, 2003)

How does Sickle Cell Disease Cause Pain?

In the oxy-haemoglobin state the sickle cell is a biconcave disc (Figure 14A). In the deoxygenated state the sickle cell changes its configuration exposing the hydrophobic sites of the tetramers, causing the molecules to form polymers (Figure 14B) that become tactoids (Figure 14C), giving rise to the 'holly leaf' configuration (Figure 14D), but when reoxygenated in the lungs the molecules return their original biconcave disc. These 'oxy' and 'deoxy' cycles result in sickling and unsickling which ultimately cause the loss of membrane material as the cells pass through the spleen. The cells become dehydrated and rigid due to leakage of K+ and water; they become less deformable and eventually become boat shaped (twice as long as they are wide; irreversibly sickled cells; Figure 14E) that are no longer able to change their shape until they are removed from circulation prematurely (Shen *et al*, 1949). In the deoxy state, haemoglobin causes the red cells to be sticky through adhesion receptors CD36 that bind to thrombospondin and integrin, which binds to fibronectin and vascular cell adhesion molecules. Then the rigid, poorly deformable red cells begin to accumulate and occlude the small vessels (Figure 14F), thus reducing blood flow to tissues. Ischaemic changes occur that may lead to death of tissue (necrosis) if the blood flow is not restored in time.

The process of reperfusion causes the release of acute phase proteins that cause pain, swelling, heat, redness and limitation of movement (the cardinal signs of acute inflammation). This is painful (vaso-occlusive) crisis (VOC), which can be precipitated/triggered by infections (e.g. malaria, bacteria or viruses), dehydration, extremes of temperature, physical exertion, injury (including surgical injury), emotional stress, idiosyncratic (peculiar to the individual) factors and pregnancy. These factors may also precipitate/trigger other forms of crises.

Clinical Features and Complications of Sickle Cell Disease

Clinical features of SCD vary between patients depending on severity (genetic) and environmental factors such as socioeconomic class, access to good medical care and compliance with preventive medications or interventions. There is no system in the body that cannot be affected from head to toe. The clinical features are divided into signs and symptoms seen in the steady state and/or crisis which may be associated with complications of the disease. The steady state, broadly speaking, is when there is no pain or any change in the blood due to therapy.

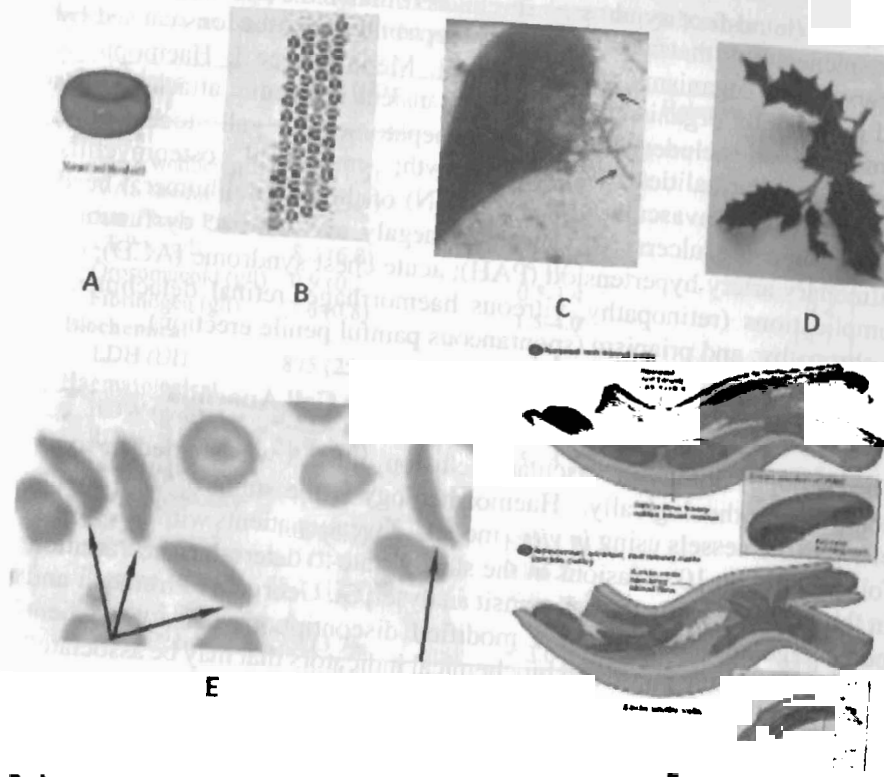

Figure 14: **A**- Oxy-haemoglobin state (biconcave disc); **B**- Polymer formation; **C**- Formation of tactoids under an electron microscope; **D**- Holly leaves representing the reversibly sickled cell configuration; **E**- irreversibly sickled cells under a light microscope; **F**- Process of vascular occlusion. (Sticky Red Cells-learning about the glue; sickle sense-wordPress.com posted May 8, 2014)

Crises, however, are punctuations in the steady state that may be associated with excruciating pain (painful crisis), hyperhaemolysis (haemolytic crisis), and worsening anaemia that may be due to sequestration crisis (pooling of blood in the reticuloendothelial system, but mainly the spleen/liver), megaloblastic crisis (deficiency of folic acid or vitamin B_{12}) or aplastic crisis (usually caused by Parvovirus B19). The common features are anaemia, lemon yellow jaundice, painful events, long thin extremities, bossing of the skull and gnathopathy (Akinyanju *et al* 1989; Akenzua *et al*., 1994). Pathological changes that occur from prolonged tissue hypoxia and infarction, result in complications, such as

dactylitis (hand-foot syndrome) in children under the age of two years; autosplenectomy that increases the susceptibility to infections caused by encapsulated organisms (Pneumococcal, Meningococcal, Haemophilus and Salmonella organisms); stroke or transient ischaemic attacks. Other complications include; splenomegaly; hepatomegaly; gall stones; other skeletal abnormalities (stunted growth; multifocal osteomyelitis; osteonecrosis or avascular necrosis (AVN) of the femoral/humeral head; and chronic leg ulcers (CLU); cardiomegaly and cardiac dysfunction; pulmonary artery hypertension (PAH); acute chest syndrome (ACD); eye complications (retinopathy, vitreous haemorrhage, retinal detachment); nephropathy; and priapism (spontaneous painful penile erection).

Painful Crisis (Vascular Occlusion) in Sickle Cell Anaemia

The pathophysiology of vascular occlusion in SCA was studied in some detail haemorheologically. Haemorrheology is the study of the flow of blood in the vessels using *in vitro* models. Twenty patients with SCA were followed up on 10 occasions in the steady state to determine the variation in the rheology using the cell transit analyser, St. George's Filtrometer and percentage of red cells on a modified discontinuous density gradient (Figure 15), acute phase and biochemical indicators that may be associated with painful crisis and acute inflammation process that ensues. The steady state was observed to be a misnomer, being found for the first time to be characterised by fluctuations (Table 2) in the percentage of poorly deformable red cells and clogging particles (rheological parameters), levels of C-reactive protein, fibrinogen and orosomucoid (acute phase) and lactate dehydrogenase (LDH; biochemical parameter) consistent with minor episodes of microvascular occlusion that were insufficient to cause overt tissue infarction of painful crisis (Akinola *et al*, 1992a).

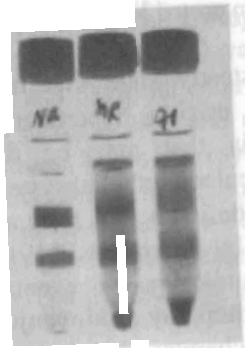

Figure 15: Modified discontinuous density gradient showing the density fractions F1, F2, F3 and F4 in Hb AA blood (NB) and two samples of Hb SS blood (MR and GI)

Table 2: Mean (SD) steady state values, reference (normal) range for apparently health adults and relative variation with time for 20 patients with SCA

Parameter	Mean (SD)	Reference Range	Relative Variation (CV %)
Clinical			
VAS wellbeing	2.0 (1.6)	10	81.9
VAS tiredness	2.7 (1.9)	0	59.8
Acute Phase Proteins			
CRP (mg/l)	8.0 (6.0)	< 10	71.6
Orosomucoid (g/l)	0.9 (0.2)	0.5-1.4	9.4
Fibrinogen (g/l)	3.6 (0.8)	1.5-4.0	14.7
Biochemical			
LDH (UI)	875 (256)	230-400	12.4
Haematological			
HDW (g/dl)	4.3 (0.5)	2.2-3.2	4.7
RDW (%)	21.9 (1.4)	11.5-14.5	3.5
Rheological			
Plasma viscosity (mPa.s)	1.69 (0.1)	1.52-1.71	4.0
MCV < 60fl (%)	6.0 (3.4)	0.1-6.7	30.5
Fraction 4 (%)	38.1 (12.7)	< 6	19.6
Clogging particles (x 10^5/ml)	9.2 (3.7)	< 3.0	24.9

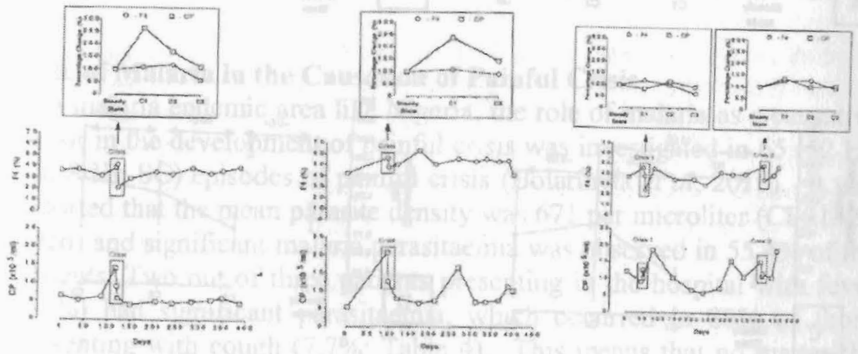

Figure 16: Examples from three patients with serial values of the percentage dense cells in F4 (%) and clogging particles (x 10^5/ml) during the steady state. Inserts show the percentage change during vaso-occlusive crisis with the mean of all the steady values expressed as 100%

Figure 16 shows the rheologic variations between patients and within a patient with time. The steady state was therefore defined as "the period free of crisis extending from at least three weeks since the last clinical event and three months or more since the last blood transfusion (intervention), to at least one week before the start of a new clinical event" (Akinola et al, 1992a). During painful crisis, a "prodromal" phase (C1), comprising the development of a sub-population of poorly deformable dense cells, was identified just before the acute phase response on days 3-5 (C2; established phase); and resolution (C3; >5 days; Figure 17; Akinola *et al*, 1992b). C-reactive protein and serum amyloid A protein have been identified to have potential value in monitoring the onset of pain and in confirming subsequent resolution of tissue ischeamia in painful crisis (Stuart *et al*, 1994).

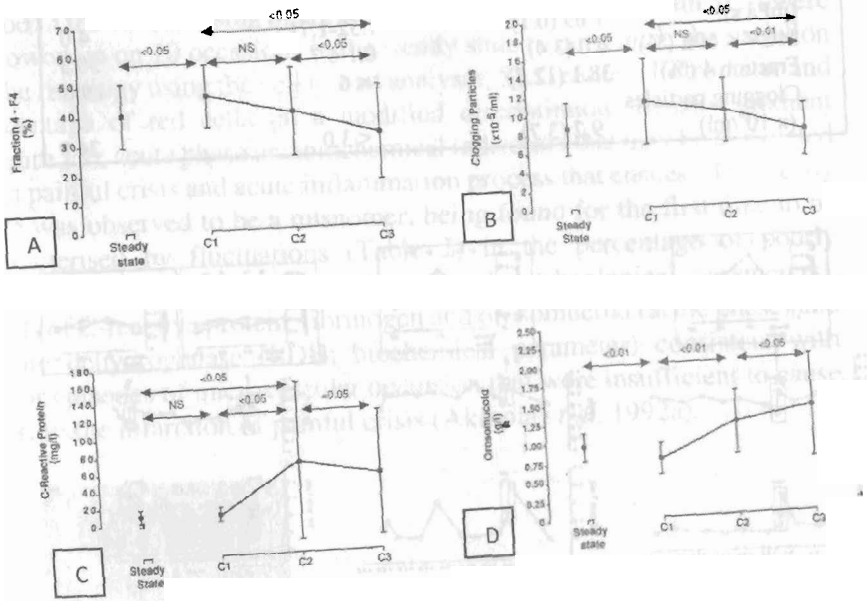

Figure 17: Comparison between mean (SD) values for percentage dense cells in F4 of the density gradient (A), clogging particles (B), C-reactive protein (C) and orosomucoid (D) in the steady state and in three stages of 12 painful crises (C1, day 1; C2, 3-5 days; C3, 6-7 days)

Abdominal Pain in Sickle Cell Anaemia

For the first time in Nigeria, abdominal pain in SCA was reported to be due to painful crisis in about one out of four patients (26%) who had Hb SS (Akinola *et al*, 2009). Other causes of abdominal pain in SCA (Table 3) were peptic ulcer disease/gastritis (28%), hepatopathy (10%), gall stones (6%), enteritis (6%) and menstruation related pain (4%). The prevalence rates and pattern of abdominal pain in Hb SS and Hb SC was similar, but abdominal crisis characterised by diffuse/generalised dull abdominal pain occurred in Hb SS patients only in this series, it was therefore regarded as a marker of severity of SCA.

Table 3: Aetiology of abdominal pain in patients with SCD

Cause of Pain	Hb SS n = 50 (100%)	Hb SC n = 8 (100%)
PUD/Gastritis	14 (28)	4 (50)
Abdominal VOC	11 (26)	0 (0)
Hepatopathy	5 (10)	1 (12.5)
Enteritis	3 (6)	2 (25)
Cholelithiasis	3 (6)	0 (0)
Menstruation related	2 (4)	0 (0)
Others	12 (24)	1 (12.5)

Key: PUD – peptic ulcer disease; VOC – vaso-occlusive crisis

Role of Malaria in the Causation of Painful Crisis

In a malaria endemic area like Nigeria, the role of malaria as a causative factor in the development of painful crisis was investigated in 65 (52 Hb SS; 8 Hb SC) episodes of painful crisis (Bolarinwa *et al*, 2010). It was reported that the mean parasite density was 671 per microliter (CI 315.9-1026) and significant malaria parasitaemia was observed in 55.4% of the patients. Two out of three patients presenting in the hospital with fever (40%) had significant parasitaemia, which occurred in 80% of those presenting with cough (7.7%; Table 4). This means that no matter the other symptoms a patient with SCD may present with in painful crisis, malaria infection should be investigated and treated as a medical emergency if positive. In addition, the effectiveness of antimalarial prophylaxis was questioned and persistent splenic enlargement was observed in 12.3% whilst enlargement of the liver occurred in 44.6% of patients in this series.

Table 4: The prevalence of clinical features in 65 vaso-occlusive episodes in relation to the percentage of malaria parasitaemia

Clinical Features	Incidence (%)	Malaria Parasite Present (%)
Pains	100	55.4
Hepatomegaly	44.6	58.8
Fever	40	61.5
Jaundice	35.4	73.9
Leucocytosis (>11.0 x 10^9/l)	35.4	43.9
Headache	23.1	53.3
Vomiting	13.8	77.8
Splenomegaly	12.3	37.5
Anaemia (PCV <18%)	12.2	75.0
Hepatosplenomegaly	9.2	50.0
Cough	7.7	80.0
Thrombocytosis (>400 x 10^9/l)	7.7	40.0
Weakness	6.2	50.0
Diarrhoea	4.6	33.3
Dysuria	1.5	0.0

Complications in Sickle Cell Disease

With advancing age, patients with SCD develop complications on account of the irreversible organ damage that may occur with increasing risk, such as cardiovascular disease, renal insufficiency and chronic lung disease.

Cardiovascular Disease in Sickle Cell Disease

My Part II dissertation for the award Fellowship (Internal Medicine) of the National Postgraduate Medical College of Nigeria was on some observations of the cardiovascular status of 22 individuals with SCA and 22 age and sex-matched controls with Hb AA at rest and in response to exercise using the modified Bruce protocol on the treadmill (**Akinola**, 1994). Some of the data obtained were presented at the 24th Annual Scientific Conference of the Nigerian Cardiac Society, Ile- Ife, Osun State, (**Akinola** and Balogun, 1995). Table 5 shows that the patients with SCA weighed less than Hb AA controls, PCV and the serum albumin of patients were also significantly less than that of controls. Patients with SCA had larger hearts (cardiomegaly) than the controls with the cardio-thoracic ratio on chest radiographs ranging from 0.46 to 0.63 in patients and 0.37 to 0.50 in controls (Table 6). The mean values of electrocardiographic (ECG) parameters showed that the mean QRS voltage was significantly higher among patients than controls and ECG abnormalities at rest, observed in 72.7%, were also more in patients than controls.

Abnormalities included left ventricular hypertrophy (LVH) by voltage criteria (63.8%), unifocal premature ventricular contractions (PVC; 13.6%) sinus arrhythmia (27.3%), prolonged PR interval (9.1%), sinus bradycardia (4.5%) and non-specific intra-ventricular conduction defect (22.7%). Specific and non-specific T-wave changes occurred more in patients with SCA than controls, but overt features of ischaemic heart disease were not seen, although Odia (1990) observed T-wave changes in more than one ventricular lead and suggested that these abnormalities may be indices of right ventricular ischaemia. Adebayo *et al.*, (2002 a) reviewed cardiovascular changes in SCA and observed that heart diseases such as ischaemic rheumatic and congenital heart diseases were rare in these individuals.

Table 5: Some clinical and laboratory features of 22 patients with SCA compared to age and sex-matched Hb AA controls

Variable	Hb SS	Hb AA	P-Value
	Mean (SD)	Mean (SD)	
Age (yr)	21.6(7.2)	22.1 (6.3)	NS
Age range	15 - 47	16 - 40	NS
M: F ratio	1:1	1.2:1	NS
Height (cm)	161.4 (10.2)	166.9 (8.7)	NS
Weight (kg)	45.6 (10.8)	55.7 (11.0)	0.0001
PCV (%)	23.1 (3.8)	39.9 (3.4)	0.0001
Serum Albumin (g/l)	35.9 (5.3)	42.4 (6.0)	<0.001

NS – not statistically significant; PCV – packed cell volume

Table 6: Cardiothoracic ratio (CTR) and electrocardiographic parameters at rest in patients with Hb SS and normal controls (Hb AA).

Variable	Hb SS	Hb AA	P-Value
	Mean (SD)	Mean (SD)	
Cardio-thoracic ratio	0.55 (0.04)	0.45 (0.03)	0.0001
CTR range	0.46 – 0.63	0.37 – 0.5	
Resting ECG Changes			
Normal: Abnormal	6:16	14:8	<0.05
Sinus rate (beats/min)	75 (13)	69 (16)	<0.05
QRS frontal axis (°)	38.6 (20.8)	47.9 (20.7)	NS
QRS voltage (mm)	45.9 (18.4)	31.5 (10.3)	<0.01
QRS voltage range	17 - 87	16 - 50	

NS – not statistically significant

Lipid Profile and Physic in Sickle Cell Disease

It is often said that individuals with sickle cell disease can be spotted from afar because of their physic, thin long extremities, but is it still true? A study of 58 patients with SCA in the steady state were studied to determine the relationship between their anthropometric parameters and lipid profile and compared with age and sex-matched Hb AA controls (Uche and **Akinola** 2017). It was observed that the weight, body mass index (BMI), mid-arm circumference, mid-thigh circumference, hip circumference, thigh length and waist/hip ratio of individuals with SCA were significantly less than those of controls (Table 7). The height, waist circumference, mid-arm and abdominal skin fold thickness were similar to those of controls. All the lipid parameters were significantly different from those of controls with the exception of triglycerides (Table 8). It was therefore concluded that although individuals with sickle cell anaemia were small in size and the mean level of total cholesterol was lower than that of controls, almost two thirds of them had normal levels contrary to expectation.

Table 7: Anthropometric data in individuals with SCA and controls

Parameter	Individuals with SCA Mean (SD) N=58	Controls Mean (SD) N=58	P-value
Height (m)	1.6 (0.1)	1.7 (0.1)	NS
Weight (kg)	52.2 (9.9)	64.4 (12.7)	< 0.0001
BMI (kg/m^2)	19.7 (3.3)	23.1 (4.5)	< 0.0001
Mid-arm circumference (cm)	25.0 (2.7)	27.1 (5.6)	< 0.05
Waist circumference (cm)	75.3 (8.3)	74.8 (10.2)	NS
Mid-thigh circumference (cm)	42.5 (5.1)	48.2 (6.1)	< 0.0001
Hip circumference (cm)	82.3 (8.2)	89.0 (10.6)	< 0.01
Mid-arm skin fold thickness (cm)	24.0 (8.3)	23.8 (9.4)	NS
Abdominal skin fold thickness (cm)	18.4 (9.2)	18.6 (8.6)	NS
Thigh length (cm)	48.8 (5.3)	51.2 (3.1)	< 0.05
Waist/hip ratio	0.92 (0.1)	0.8 (0.1)	< 0.05

NS – not statistically significant

Table 8: Lipid profile of patients with SCA compared to normal controls

Parameter	Individuals with SCA Mean (SD) N=58	Controls Mean (SD) N=58	P-value
Total cholesterol (TC; mmol/l)	3.3 (0.6)	4.0 (0.9)	< 0.0001
High density lipoproteins (HDL; mmol/l)	0.7 (0.3)	1.1 (0.8)	< 0.0001
Triglycerides (Tg; mmol/l)	1.1 (0.5)	0.8 (0.3)	0.078*
Low density lipoproteins (LDL; mmol/l)	2.1 (0.5)	2.7 (0.9)	< 0.01
Non-HDL (mmol/l)	2.5 (0.6)	2.8 (0.9)	< 0.05
TC/HDL ratio	4.5 (1.4)	3.7 (1.2)	< 0.05

* not statistically significant

The risk factors for ischaemic heart disease such as obesity, high levels of cholesterol, atheroma and high blood pressure are uncommon in SCD (Tables 7 and 8). The mean body mass index (BMI) of the patients studied (Uche and **Akinola**, 2017) was normal, but significantly lower (19.7±3.3 kg/m^2) than age and sex-matched Hb AA controls (23.1±4.5 kg/m^2); the mean total cholesterol in serum (3.3±0.6 mmol/L) for patients was significantly lower than that of controls (4.0±0.9; $P < 0.0001$). At rest, the mean heart rate/pulse rate was normal, but significantly higher than that of controls whilst the mean blood pressure was lower, but not significantly different from that of controls (Table 9). Exercise duration was significantly reduced among patients [375.8 (81.5) sec] than controls [540.1 (84.4) sec], maximum heart rate, change in heart rate and heart rate five minutes post exercise were significantly less than those of controls. However, the systolic and diastolic blood pressures were not significantly different from those of controls, but the mean pulse pressure was significantly higher than that of controls. Exercise capacity, MO_{2max}, was also significantly reduced in patients than controls.

Cardiac complications of SCA was studied further in a cohort of 41 patients and age and sex-matched Hb AA controls using self-paced walking exercise testing (Adebayo, *et al.*, 2002b) and echocardiography (Adebayo, *et al.*, 2004) in the steady state. Attention was paid to the non-specific ECG changes that included, a QRS voltage by Sokolow-Lyon criteria that was significantly higher in patients with SCA than normal controls ($P < 0.05$). The mean resting heart rate, P-wave duration and corrected QT interval were significantly higher in patients than controls.

Table 9: Some haemodynamic parameters of patients with SCA compared to normal controls at rest, during exercise and five minutes after exercise.

Variable	Hb SS	Hb AA	P-Value
	Mean (SD)	Mean (SD)	
Resting			
Heart rate (beats/min)	78 (12)	69 (15)	<0.05
SBP (mmHg)	94.8 (12.8)	99.6 (12.0)	NS
DBP (mmHg)	55.9 (11.4)	60.5 (8.5)	NS
During Exercise			
Duration (sec)	375.8 (81.5)	540 (84.4)	0.0001
Max HR (mmHg)	167 (25)	186 (27)	<0.05
Max SBP (mmHg)	129.1 (17.4)	136.8 (16.7)	NS
Max DPB	73.2 (7.8)	77.3 (7.0)	NS
MO_2 max	29153 (4700)	25361 (4592)	<0.05
Change in HR	88 (26)	116 (24)	<0.001
Change in SBP	34.3 (15.6)	37.3 (13.1)	NS
Post Exercise			
HR (beats/min)	90 (10)	99 (12)	<0.05
HR range	72 -104	84 - 128	
SBP (mmHg)	97 (10)	104 (12)	NS
SBP range	80 - 120	90 - 130	

NS – not statistically significant; SBP – systolic blood pressure; DBP – diastolic blood pressure

A significantly lower mean QRS frontal axis was observed in patients when compared to controls ($P< 0.05$), but there was no significant difference in the mean QRS duration and PR interval between the two groups. No abnormal QRS axis was seen in either group, therefore ischaemic heart disease is rare among patients with SCA and young adults with Hb AA. Self-paced walking exercise showed that patients with SCA significantly had limited exercise capacity when compared with controls evidenced by the reduction in the distance covered and speed achieved. Although patients had a significantly smaller change in heart rate, post exercise heart rate and systolic BP were similar to those of controls. It was therefore concluded that exercise capacity was limited in self-paced walking exercise, which was found to be a safe and reproducible measure of cardiac reserve in these patients. Non-invasive assessment of cardiac function in patients with sickle cell anaemia (Adebayo, *et al.*, 2004) showed that they had significantly higher left ventricular (LV), right ventricular (RV), and left atrial (LA) dimensions and LV mass, but the ejection phase indices of the LV systolic function were similar to those of

controls. Trans-mitral Doppler indices of LV diastolic filling were, however, significantly less than in controls. This therefore implies that left ventricular diastolic function was impaired in some patients despite normal systolic function.

Pulmonary Artery Hypertension

Sickle cell disease is associated with sudden death (Gladwin et al, 2004) and reduced live expectancy (Platt et al, 1994) and pulmonary artery hypertension (PAH) has been found to play a role. Pulmonary artery hypertension is defined as the mean pulmonary artery pressure greater than 25 mmHg at rest, measured during right heart catheterization. In the absence of cardiac catheterization, pulmonary pressures may be derived from measurements on echocardiography or Doppler-echocardigraphy. A consensus guideline has recently been published by the American Thoracic Society and endorsed by the Pulmonary Hypertension Association and the American College of Chest Physicians (Klings, *et al.*, 2014). These guidelines summarize that an increased risk for mortality is associated with increased TRV $\geq$ 2.5 m/s, right heart catheterization confirmation of PAH with a mean pulmonary artery pressure $\geq$ 25 mmHg (either PAH or PVH), and an elevated NT-BNP $\geq$ 160 pg/ml. Stable SCD patients should be screened with Echo and/or NT-BNP every 1–3 yr to assess this mortality risk. If patients are found to have TRV > 2.5 m/s and or NT-BNP > 160 pg/ml, suggesting an increased risk of PAH and higher mortality, they should undergo right heart catheterization to confirm the diagnosis of PAH and guide specific therapy (Klings *et al.*, 2014). The few studies in literature showed that PAH occurred variably in up to 30% of patients with SCD using echocardiography-based techniques (Castro et al., 2003; Gladwin, et al., 2004). Aliyu, *et al.*, (2008) reported the prevalence of PAH as 25% amongst Nigerian patients using peak TRV of $\geq$2.5 m/s as a criterion, whereas, Dosunmu, *et al.*, (2014) in a study in Lagos, Nigeria, obtained a much lower prevalence of 3.6%. The difference in these two studies may be due to technique and the inclusion of younger age groups. To determine the prevalence in adults, Amadi, *et al.*, (2017) at OAUTHC, Ile-Ife, Nigeria, screened 92 patients with SCA for PAH and assessed for the influence of PAH on exercise capacity (6-minute self-paced walk) and determined the correlates and predictors of the estimated pulmonary pressure. Diagnosis of PAH was by Doppler echocardiography on finding a tricuspid regurgitant velocity (TRV) of $\geq$ 2.5 m/s and the pulmonary flow profile was also assessed to estimate the mean pulmonary arterial pressure (MPAP).

Table 10: Relationship between PAH and clinical parameters

Parameter	PAH	No PAH	*p* value
Age (years)	28.6 (5.8)	23.4 (3.4)	<0.001*
PCV (%)	25.6 (2.9)	27.5 (2.4)	0.001*
HR (beats/min)	84.0 (8.2)	83.3 (3.7)	0.616
SBP (mmHg)	107.5 (12.1)	105.5 (8.9)	0.264
DBP (mmHg)	64.5 (10.2)	64.5 (8.2)	0.987
BMI (kg/m^2)	19.1 (2.6)	18.9 (1.4)	0.810
BSA (m^2)	1.50 (0.15)	1.53 (0.14)	0.482
6MWD (m)	380.3 (63.2)	474.3 (76.7)	0.014*

*Statistically significantly different

Abbreviations: PAH, pulmonary artery hypertension; PCV, packed cell volume; HR, heart rate; SBP, systolic blood pressure; DBP, diastolic blood pressure; BMI, body mass index; BSA, body surface area; 6MWD, 6-minute walk distance.

Doppler-derived PAH was detected in 23.9% of adults with SCA using TRV and 38% using pulmonary flow Doppler. The 6-minute walking distance (6MWD) was significantly lower in SCA adults with PAH than in those without PAH (380.33±63.17 m vs 474.28 ±76.74 m; $p = 0.014$). TRV and estimated MPAP had a significant inverse correlation with the 6MWD (r= -0.442; $p < 0.001$ and r = -0.571; $p < 0.001$, respectively). It was concluded that older patients with lower haematocrit are at risk of PAH and they should be investigated and treatment early to prevent sudden death and thus improve their quality of life and life expectancy.

Table 11: (A) Significant correlates of estimated MPAP and (B) independent parameters linked to mean pulmonary arterial pressure

A

Parameter	Pearson's correlation	*P*- value
Age (years)	0.467	<0.001
Haematocrit (%)	–0.467	<0.001
6MWD (m)	–0.571	<0.001
LADi (cm/m^2)	0.397	<0.001
LVEF (%)	–0.257	0.013
TDe2 (cm/s)	–0.322	0.002
TDs2 (cm/s)	–0.387	<0.001
E/*e*′ ratio	0.310	0.002
RVIDDi (cm/m2)	0.249	0.016

B

Parameter	Unstandardized coefficient	Standardized coefficient	*P*-value
Haematocrit (%)	–1.105	–0.270	0.003
Age (years)	0.678	0.315	0.004
LADi (cm/m2)	10.283	0.288	0.004
6MWD (m)	–0.041	–0.231	0.038
Constant	67.237		

Note: TDe2, early mitral annular septal tissue diastolic velocity; TDs2, mitral annular septal tissue systolic velocity; *E*/*e*2, ratio of early transmitral diastolic flow velocity to early mitral annular septal tissue diastolic velocity.
Abbreviations: MPAP, mean pulmonary arterial pressure; 6MWD, 6-minute walk distance; LADi, left atrial dimension index; EF, left ventricular ejection fraction;

Renal Complications

The kidney is another essential organ of the body which was studied in individuals with SCD. It is known that kidney function deteriorates with age and is associated with morbidity and mortality. The kidney in SCD is affected by both the haemodynamic changes of chronic anaemia and the consequences of recurrent vaso-occlusion leading to structural and functional changes and progression to chronic kidney disease (CKD; Serjeant and Serjeant 2001; Kadiri 2006). Sickle cell nephropathy is a spectrum of morphologic, laboratory and clinical changes characterized by specific manifestations, risk factors, and prognostic indicators that include polyuria, haematuria, proteinuria, renal failure syndromes and distinct glomerular or tubular lesions (Walker *et al.*, 1971; Phuong-Thu *et al.*, 2000). It is associated with a markedly increased risk of mortality with little in the way of specific therapy available to increase life expectancy. Proteinuria, hypertension, severe anaemia, and haematuria were found to be reliable predictors, while the inheritance of the CAR haplotype increased the risk (Powars *et al.*, 1991; Platt *et al.*, 1994).

The burden of sickle cell nephropathy is high among Nigerian patients and the process of collecting urine over 24 hours for the measurement of creatinine clearance to determine kidney function is often associated with inherent errors. The patients with SCD at OAUTHC, Ile-Ife, Nigeria were therefore studied to evaluate formulae that have been commonly used to predict the magnitude of kidney dysfunction together with predictive factors (Arogundade, *et al.*, 2011; Adegoke, *et al.*, 2012; Bolarinwa, *et al.*, 2012; Aneke et al., 2014; Aneke *et al.*, 2015). In the Arogundade series,

data from 374 patients with SCD were reported between the ages of 7 and 62 years (median 23 years), while the median age at diagnosis of SCD was 4 years (range 3 months - 31 years). No kidney disease was observed in 235 (68.2%) patients, while the remaining 139 (37.2%) had proteinuria, haematuria or reduced glomerular filtration rate (GFR) < 60 ml/min. The age of patients was a significant predictor of kidney disease ($p = 0.002$) and correlated with the level of serum creatinine ($r = 0.188$, $p < 0.001$), GFR ($r = 0.245$, $p < 0.0001$) and the degree of proteinuria ($r = 0.174$, $p = 0.006$). Patients with kidney disease had a significantly higher number of crises/hospitalizations ($p < 0.001$). Seven (1.87%) patients died of whom 4 (57%) had end-stage renal disease. It was concluded that kidney disease is a common complication of SCD and significantly contributes to mortality. The age of the patients, that is, the duration of SCD and frequency of crises/hospitalizations (determinants of severity) were strong predictors of the development of kidney disease.

Table 12: Demographic and clinic characteristics of study

Parameter	SCD (n = 100)	HbSS (n = 79)	HbSC (n = 21)
Age (years)	26.2 ± 7.4	25.3 ± 6.7	29.3 ± 8.9
Weight (kg)	51.9 ± 10.47	49.4 ± 9.0	61.2 ± 11.7
BMI (kg/m^2)	19.5 ± 3.9	18.3 ± 2.5	23.7 ± 5.1
Serum creatinine (μmol/ml)	83.0 ± 22.1	82.0 ± 20.4	88.7 ± 27.8
Urine creatinine (μmol/ml)	3949 ± 2190	3813 ± 2120	4457 ± 2422

Table 13. Comparison of GFR methods in study population (n = 100)

Methods	Mean	SD	P-value
Measured Creatinine clearance (ml/min)	66.80	26.36	-
Gates (ml/min/m^2)	66.24	28.31	0.9
Cockroft-Gault (ml/min/m^2)	56.40	16.68	< 0.001
Mawer (ml/min/m^2)	58.38	17.47	< 0.01
Hull (ml/min/m^2)	81.98	27.48	< 0.001
MDRD (ml/min/m^2)	115.15	39.84	< 0.001

In an attempt to find an alternative means of assessing the status of renal function in patients with SCD, Adegoke *et al.*, (2012) for the first time in Nigeria, evaluated five predictive formulae in these patients by comparing them with the 24 hour urine estimation of glomerular filtration rate obtained. One hundred SCD patients (79 Hb SS and 21 Hb SC) were studied (Table 12). The mean age was 26.2 ± 7.4 years and 54% were females (Table 12). Table 13 shows a comparison between measured and estimated GFR using five formulae. The highest agreement was between measured GFR and Cockcroft-Gault (C-G) estimates ($k = 0.50$), followed by Mawer ($k = 0.49$), Hull ($k = 0.26$), Gates ($k = 0.21$) and MDRD ($k = 0.02$). Using the Bland-Altman technique, the Hull (mean = -15 ± 55) and MDRD (mean = -48 ± 61) formulae significantly underestimated GFR while the Mawer (mean = 8 ± 39) and CG (mean = -10 ± 39) formulae overestimated GFR. The Gates formula (mean = 0.6 ± 54) showed no difference with measured GFR. It was concluded that the predictive formulae commonly used to assess the status of renal function may cause inappropriate classification of many patients with SCD. It was suggested that the Cockcroft–Gault formula may be used with the understanding of its limitation.

Table 14: The *stages of kidney disease according to the eGFR (mL/min/1.73 m^2) of the study subjects

Stage of kidney disease	Glomerular filtration rate (eGFR)	Number of patients involved	Percentage of patients in the group with albuminuria
Hyperfiltration	>140	22 (30.6%)	36.4
Stage 1	90–140	18 (25.0%)	44.4
Stage 2	60–89	30 (41.7%)	73.3
Stage 3	30–59	2 (2.7%)	100
Stage 4	15–29	Nil	-
Stage 5	<15	Nil	-

*International classification of chronic kidney disease based on the clinical estimation of GFR (eGFR) by Skorecki, *et al.*

Bolarinwa, *et al.*, (2012) determined the prevalence, pattern and the associated risk factors of renal disease in 72 adults with SCA from two centers in the southwestern Nigeria. The presence of albuminuria of at least 1+ or microalbuminuria in those negative with dipstick; and the estimated glomerular filtration rate (eGFR) using the Cockcroft–Gault (C-G) formula were used to categorize patients into the various stages of chronic kidney disease (CKD). The eGFR showed that 22 (30.6%) patients

had hyperfiltration (GFR > 140 mL/min/1.73 m2), of whom 36.4% had albuminuria, 18 (25.0%) had stage 1 CKD, 30 (41.7%) had stage 2 CKD and two (2.7%) patients had stage 3 CKD with albuminuria, while none had stages 4 and 5 CKD (Table 14). Patients with and without albuminuria were compared to determine the relative risk associated with renal disease. Four (5.6%) patients had macro-albuminuria, while 32 (44.4%) had micro-albuminuria and 30 (41.7%) had haemoglobinuria. In the patients with albuminuria, age, haematocrit, systolic blood pressure, serum creatinine, urea, creatinine clearance and eGFR were not significantly different. Only the diastolic blood pressure was significantly higher in the albuminuric group (Table 15). It was therefore concluded that renal abnormalities, importantly albuminuria, is common in adult Nigerians with SCA and the pattern and incidence are similar to those reported from other parts of the world. Regular blood pressure monitoring, early diagnosis and active intervention are advocated to delay progression to end-stage kidney disease in view of the poor outcomes of renal replacement therapy in SCA patients with nephropathy.

Table 15: The characteristics and laboratory data among non-albuminuric and albuminuric subjects

Characteristics	Non-albuminuric subjects (n=40) Mean ± SD	Albuminuric Characteristics subjects (n=28) Mean ± SD	*P*-value
Age (years)	23.6 ± 10.6	25.1 ± 9.5	NS
Haematocrit (%)	23.6 ± 2.7	24.0 ± 3.5	NS
WBC count (mm/mL)	12,943 ± 3,796	12,812 ± 4120	NS
VOC/year	3.8	4.3	NS
Transfusion/year	2.2	1.8	NS
SBP (mmHg)	111.8 ± 13.1	112.5 ± 9.1	NS
DBP (mmHg)	63.1 ± 12.9	78.3 ± 17.1	0.043*
Body mass index (kg/m^2)	19.4 ± 4.3	18.1 ± 2.7	NS
Blood urea nitrogen (mmol/L)	2.5 ± 0.7	3.2 ± 0.8	NS
Serum Creatinine (µmol/L)	62.3 ± 30.8	86.8 ± 44.4	NS
Creatinine Clearance (mL/min)	99.0 ± 27.1	97.5 ± 29.2	NS
GFR (mL/min/1.72 m^2)	126.3 ± 46.7	103.7 ± 54.3	NS

NS – not statistically significant; VOC – vaso-occlusive crisis; SBP – systolic blood pressure; DBP – diastolic blood pressure;

Aneke *et al.*, (2014) determined the degree of chronic kidney disease (CKD) using creatinine clearance in 100 (79 Hb SS and 21 Hb SC) adult Nigerian patients with SCD and 50 Hb AA controls. Table 16 shows the distribution of kidney disease categorized into stages of CKD. Of the 79 patients with Hb SS, 14 (18%), 28 (35%), 33 (42%) and 4 (5%) had stage 1, 2, 3 and 4 CKD, respectively; while the Hb SC group, 3 (14%), 9 (43%) and 9 (43%) patients had stage 1, 2 and 3 CKD, respectively. Proteinuria was noted in 16 (20%) Hb SS patients, but not in any of the patients with Hb SC. Of the patients aged between 16 and 24 years (n = 49), 9 (18%), 18 (37%), 21 (43%) and 1 (2%) had stage 1, 2, 3 and 4 CKD, respectively. Of those aged >24 years (n = 51), 8 (16%), 19 (37%), 21 (41%) and 3 (6%) had stage 1, 2, 3 and 4 CKD, respectively (Table 16). None of the subjects had stage 5 CKD. It was concluded that adult patients with SCD had variable degrees of CKD. Adequate follow-up and active intervention are advocated to delay the onset of end-stage nephropathy.

Table 16: Characteristics of all HbSS and HbSC patients

	Total	Stage 1	Stage 2	Stage 3	Stage 4	Stage 5
Hb SS patients	79 (100)	14 (18)	28 (35)	33 (42)	4 (5)	0 (0)
Proteinuria	16 (20)	2 (13)	3 (19)	7 (44)	4 (25)	0 (0)
No proteinuria	63 (80)	12 (19)	25 (40)	26 (41)	0 (0)	0 (0)
Hb SC patients§	21 (100)	3 (14)	9 (43)	9 (43)	0 (0)	0 (0)
Age						
<24 years	49 (100)	9 (18)	18 (37)	21 (43)	1 (2)	0 (0)
>24 years	51 (100)	8 (16)	19 (37)	21 (41)	3 (6)	0 (0)

[the number of patients (%); §None of the Hb SC subjects had proteinuria.]

Aneke *et al.*, (2015) went further to compare measured GFR with eGFR using the C-G and CKD-EPI formulae in patients Hb SS. The C-G formula had a stronger correlation coefficient of r = 0.667 (P < 0.001) than CKD-EPI formula (r = 0.598; P < 0.001). Whereas, among patients with Hb SC, C-G formula had a comparable correlation coefficient of r = 0.819 (P < 0.001) with CKD-EPI formula (r = 0.848; P < 0.001; Table 17). It was therefore concluded that the CKD-EPI formula is a good estimate of GFR, but it did not perform significantly better than the C–G formula in this cohort of patients.

Table 17: Correlation of measured and calculated glomerular filtration rate (GFR) in HbSS and HbSC

Subjects	Measured GFR/CKD-EPI R-value (P-value)	Measured GFR/C-G R-value (P-value)
Hb SS (n = 79)	0.598 (< 0.001)	0.667 (< 0.001)
Hb SC (n = 21)	0.848 (< 0.001)	0.819 (< 0.001)

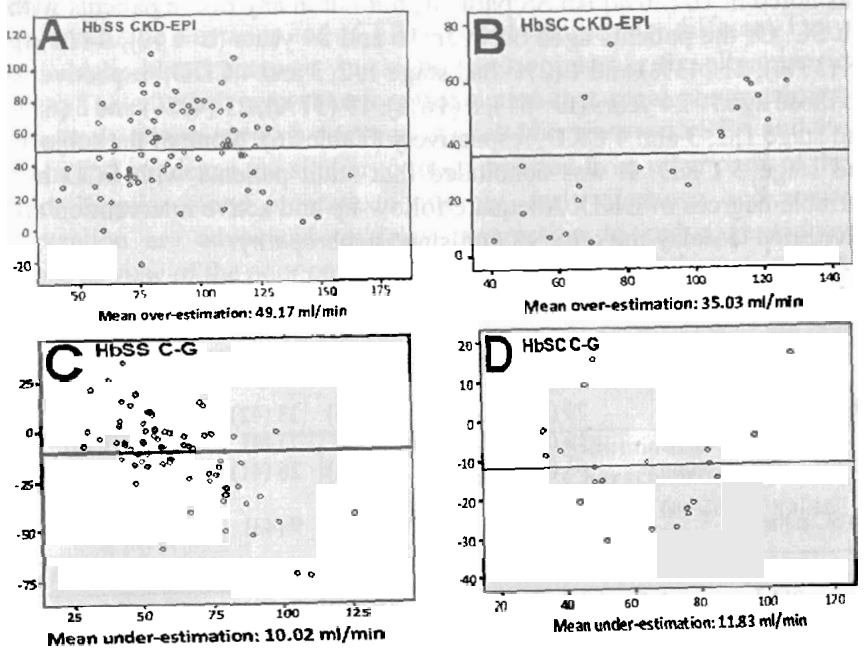

Figure 18: Bland–Altman analysis of measured versus estimated glomerular filtration rate (eGFR) using CKD-EPI and Cockcroft–Gault (C–G) formulae. The Bland–Altman plots for heterozygous haemoglobin SC (b, d) and homozygous haemoglobin SS (a, c) show overestimation of the measured GFR by the CKD-EPI formula (a, b), whereas the C–G formula underestimates the measured GFR (c, d).

Avascular Necrosis and Quality of Life in Sickle Cell Disease

Survivors of SCD face the reality of living with a chronic illness with attending physical and psychological complications that are accompanied by pain, apprehension, difficulty with fulfilling personal and family obligations, stigma and the fear of the unknown, all of which can affect quality of life (Muldoon *et al.*, 1998). A modified version of the WHO Quality of Life Brief version (WHOQOL-BREF) questionnaire was used

to assess 80 adult patients and age and sex-matched apparently healthy volunteers served as controls at OAUTHC, Ile-Ife (Mosaku *et al.*, 2015). Significantly fewer individuals with SCD were married compared to the controls (Table 18; $P = 0.01$) and their clinical characteristics are presented in Table 19.

Table 18: Socio-demographic variables

Variables	SCD	Controls	P-value
Age (years; mean (SD)	25.7 (7.7)	26.2 (5.8)	0.64
Sex			
Male (%)	33 (41.3)	33 (41.3)	<0.001
Female	47 (58.7)	47 (58.7)	1.00
Education			
Primary	6 (7.5)	4 (5.0)	
Secondary	20 (25.0)	34 (42.5)	
Tertiary	54 (67.5)	42 (52.5)	0.06
Marital Status			
Married	15 (18.8)	28 (35.0)	
Single	62 (77.5)	44 (55.0)	
Divorced/separated /widowed	3 (3.9)	8 (10.0)	0.01
Total	80	80	

Key: P value = level of significance; SD=Standard deviation, SCD = Sickle cell disease

Table 19: Clinical characteristics of participants with SCD

Clinical Features	Yes (%)	No (%)
Jaundice	44 (56)	35 (44)
Leg ulcers	12 (16)	65 (84)
Avascular necrosis	13 (19)	56 (81)

Blood transfusion in 1 year	No.	0	1-2	3-4	≥5
	79	56	20	1	2
Bone pain in 1 year	No.	0	1-3	4-6	≥7
	80	15	47	10	8
Bone pain associated with menstruation in 1 year	No.	0	1-3	4-6	≥7
	46	26	14	4	2
Priapism in 1 year	No.	0	1-5	6-10	>10
	31	21	5	2	3

Similarly, individuals with SCD scored significantly less in the physical and psychological domains as well as in overall QoL and general health domains when compared to controls (F = 0.001; Table 20). Avascular necrosis of the femur significantly affected the overall QoL and general health of these individuals with SCD, respectively while the means of the QoL assessment domains were not significantly different in individuals with SCD with and without complications, except in the general health domain (F < 0.00; Table 21). It was concluded that vascular necrosis of the femoral head significantly affects the overall QoL of individuals with SCD.

Table 20 Comparison of mean score on transformed WHO QOL BREF domains

Domains	SCD	Controls	T-test	P-value (95% CI)
Physical	13.7 (1.9)	15.5 (1.3)	6.9	0.0001(1.29-2.31)
Psychological	14.0 (2.0)	16.2 (1.8)	2.2	0.0001(1.66-2.31)
Social	15.0 (2.7)	15.5 (2.1)	1.31	0.19 (0.2-1.26)
Environmental	14.1 (2.0)	15.0 (1.8)	2.58	0.01 (0.21-1.55)
Overall QOL score	3.32 (0.78)	4.2 (0.91)	6.57	0.001 (0.62-1.14)
General health	3.5 (1.0)	4.1 (1.0)	3.59	0.0001 (0.27-0.9)

Significant P-value is shown in bold type, QOL=Quality of control, CI=Confidence interval, SCD=Sickle cell disease, SD=Standard deviation

Table 21: Comparison of means of quality of life (QOL) domains and SCD-related complications

Domains	SCD complications		T-test	P-value
	Mean (SD)			(95% CI)
	Yes	No		
Physical	12.80 (1.96)	14.2 (1.81)	4.69	0.01 (0.80-1.98)
Psychological	13.40 (2.00)	13.92 (2.30)	1.53	0.13 (0.15-1.20)
Social	14.98 (2.71)	15.35 (2.53)	1.25	0.21 (0.29-1.33)
Environmental	14.11 (2.65)	14.14 (2.35)	0.08	0.94 (0.75-0.81)
Overall QOL score	4.0 (0.80)	4.5 (0.82)	3.49	0.0001(0.22-0.78)
General health	3.25 (0.10)	3.7 (0.80)	4.99	<0.001(0.27-0.63)

Significant P-value is shown in bold type, QOL=Quality of control, CI=Confidence interval, SCD=Sickle cell disease, SD=Standard deviation

Severity of Sickle Cell Disease

The severity of SCD depends largely on the genetic inheritance and interactions that influence the production of Hb F. The higher the percentage of Hb F (Arab-India Haplotype) the milder the disease. The compound states of SCD other than Hb Sβ^0 thal (e.g. Hb SC, SD, SE etc.) tend to ameliorate the severity of the disease resulting in a milder clinical course (Driscoll 2007; Adekile 2009). Factors indicative of severe disease are associated with a low Hb F level (e.g. Benin haplotype) and include frequent painful crisis (> 3 per year); frequent blood transfusion (> 3 per year); severe anaemia (PCV < 18%); an absolute neutrophil count ≥ 10 x 10^9/l in the steady state; platelet count ≥ 500 x 10^9/l in the steady state; an abnormal TCD velocity (≥ 180 cm/sec) in children below 17 years; acute chest syndrome (ACS; characterised by dyspnoea with or without fever, increased pulse and respiratory rates, cyanosis, pulmonary signs and new infiltrates on chest X-ray bilaterally and a progressively falling SpO2 below normal); and stroke (FMOH 2014).

Haematological Profile and Disease Severity in Sickle Cell Anaemia

The severity of the SCD determines the mortality of the patient, as those with severe disease tend to suffer early organ damage and die at a relatively younger age. The median life expectancy estimated to be 42 years for males and 48 years for females (Platt et al., 1998) in the United States of America, while in Nigeria patients with SCA now survive beyond the 4th decade with optimal care (Chijioke A, Kolo PM. 2009). Clinical severity of SCA is variable and poses a challenge to the physician and families of affected persons, therefore the disease severity scores available were modified and tested against the haematological profile of patients with SCA in the steady state (Table 22; Owojuyigbe 2011).

Vascular Phenotypes of Sickle Cell Disease

Kato *et al* (2007) categorised sickle cell disease into two, the viscosity vaso-occlusive subphenotype and the haemolytic-endothelial dysfunction subphenotype. The viscosity vaso-occlusion subphenotype consists of VOC, AVN and ACS is characterised by low haemolytic rate, high haemoglobin level; higher plasma arginine and higher NO bioavailability. The haemolysis-endothelial dysfunction sub-phenotype include CLU, Priapism, PAH and may be stroke. This sub-phenotype is characterised by higher free plasma haemoglobin and arginase, higher reticulocyte count, higher serum LDH, and higher bilirubin than the viscosity VOC sub-phenotype.

Table 22: Modification of three existing scoring systems for assessing disease severity in SCD

S/N	PARAMETER	AKENOVA ET AL	BIENZLE ET AL	DIOP ET AL	MODIFIED SCORING SCHEME SCORE IN BRACKETS		
					MILD	MODERATE	SEVERE
1	NO. OF CRISES PER YEAR	+	–	+	< 2 (1)	2-3 (2)	>3 (3)
2	STEADY STATE PCV	+	+	–	> 24% (0) 21–23.5% (1)	18 – 20.5 (2)	<18% (3)
3	NO. OF BLOOD TRANSFUSION PER YEAR	–	+	+	< 2 (1)	2-3 (2)	>3 (3)
4	NO. OF HOSPITAL ADMISSIONS PER YEAR	–	+	–	< 2 (1)	2-3 (2)	>3 (3)
6	NO. OF ORGANS INVOLVED (Add scores from Table 2)	+	+	+	0-1 ORGANS (≤ 2)	2-3 ORGANS (3-6)	>3 ORGANS (≥ 6)
7	WBC COUNT (x 10^9/L)				<11 (0)	11 –15 (1)	>15 (2)
8	SUBJECTIVE ASSESSMENT (VISUAL ANALOGUE SCORE)	+	–	+	< 2 (1)	2-3 (2)	>3 (3)
	POSSIBLE SCORE				7	8 17	≥ 18

Laboratory Investigations for Sickle Cell Disorder

There are many tests that may be used to screen for and diagnose sickle cell disorder. The screening tests (Figure 19) include; peripheral blood film (A), sickling test (Na+ metabisulphite; B), solubility test (Na+ dithionite; C) and point of care test kits (Monoclonal antibody test; SickleSCAN™ /Hemo SC; Figure 19).

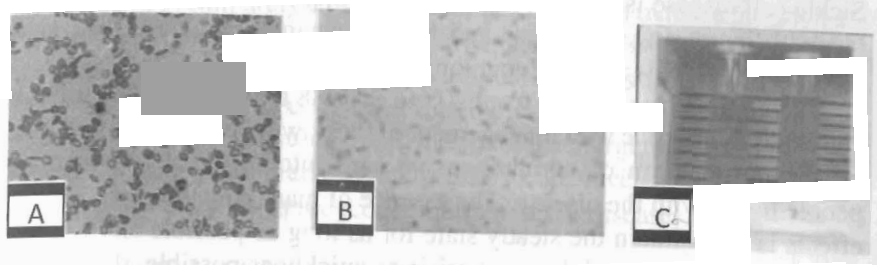

Figure 19: Peripheral blood film (A) the sickling test (B) and solubility test (C)

Diagnostic tests usually by electrophoresis include cellulose acetate (Figure 20; pH 8.4); agarose gel (pH 6.0); iso-electric focusing (IEF); and high performance liquid chromatography (HPLC). Cellulose acetate and agarose gel methods are preferably used after the age of six months when the majority of the haemoglobin in blood is the usual adult haemoglobin. High performance liquid chromatography and iso-electric focussing are ideal for newborn screening programmes. The results from these tests are called **PHENOTYPES**.

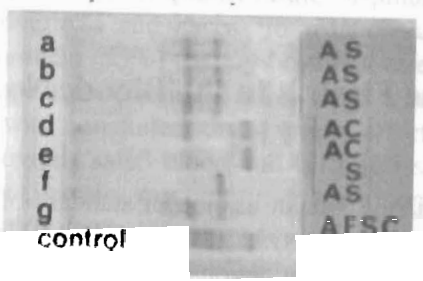

Figure 20: Haemoglobin electrophoresis using cellulose acetate at pH 8.4.

Confirmatory test is the DNA finger printing which determines the haplotype of the sickle cell gene inherited. This is DNA analysis that confirms the diagnosis of SCD; it is the **GENOTYPE**. This method is used for prenatal diagnosis (PND) and pre-implantation genetics.

Treatment of Sickle Cell Disease

Sickle cell disease is a major cause of morbidity and mortality in Nigeria, but with increasing awareness and better management strategies, many affected individuals are surviving longer. Akinyanju *et al.*, (2005) reported a reduction in mortality rate from 20.6% in 1988 to 0.6% in 1995 and this continues to improve with management of the down-stream effects of SCD by the introduction of comprehensive care into the treatment plan of people living with the disease. The essence of managing the down-stream effects is to maintain the steady state for as long as possible and remove the precipitating factor/trigger of crisis as quickly as possible, if possible, while providing medical interventions to restore the steady state. This means that regular monthly or three monthly visits to the sickle cell clinic in the steady state is necessary for clinical evaluation particularly of the level of blood, using the packed cell volume (PCV), which is cheap and readily available, size of the spleen or the liver and the presence or absence of albumin in urine. To maintain the steady state routine prophylactic measures should be administered. This includes the prevention of malaria, use of folic acid daily and prevention of infection in children under the age of 17 years either by immunisation or with the use of Penicillin V tablets. Whenever there is crisis the patient should be referred to a secondary or tertiary health facility where necessary interventions like intravenous fluids, blood transfusion and appropriate antibiotic regimens can be administered.

"The Life of the Flesh is in the Blood; No Blood No Life"

Since there is still variably poor utilisation of standard-of-care practices for patients with SCD in Nigeria, OAUTHC, Ile-Ife, took part in a survey to assess the common management options and facilities available in some dedicated SCD clinics in Nigeria (Galadanci *et al.*, 2013). Responses were obtained from 18 clinics based in 11 health facilities nationwide. All the facilities had electronic cell counters exce three, but all had access to haemoglobin electrophoresis. Three ha high-performance liquid chromatography (HPLC) machines instal d, but none was being used

routinely. Only one institution had a functioning molecular biology laboratory and an official newborn screening programme was not available in the country at the time of the survey. All facilities had access to microbiology and chemistry laboratories, but nine (81.8%) facilities had CT while six (54.5%) had MRI and three (27.3%) had transcranial Doppler facilities. The number of patients being followed in each centre ranged from 15 to approximately 11,000. All clinics provided malaria prophylaxis and folic acid routinely to patients. Only eight (44.4%) clinics prescribed penicillin prophylaxis and hydrocarbamide to patients who could afford it when indicated. It was concluded that the care available to patients with SCD in Nigeria is still suboptimal and there is an urgent need to improve quality of care, particularly at the primary care level, where significant impact could be made to reduce the burden of the disease.

Blood Transfusion in Sickle Cell Disease

Safe, timely red blood cell transfusion saves lives and chronic transfusion therapy (CTT) prevents or limits morbidity and mortality in patients with SCD, thus improving quality of life (Diaku-Akinwumi, *et al.*, 2016). This multicentre questionnaire-based study assessed the ability of hospitals in Nigeria to provide "safe" blood to patients with SCD between March and August 2014. Thirty one of the 73 hospitals contacted responded, but 24 (78%) hospitals were unable to transfuse patients regularly due to blood scarcity. Packed red blood cells were available in 14 (45%), while only one provided leukocyte-depletion. Most hospitals assessed donor risk and screened for HIV (30; 97%), hepatitis B (31; 100%) and hepatitis C (27; 87%). Extended phenotyping and alloantibody screening were not available in any hospital. A quarter of the hospitals used serum ferritin to monitor iron overload and access to iron chelators was limited and expensive. Seventeen (55%) tertiary hospitals offered CTT by top-up or manual exchange transfusion; previous stroke was the most common indication. It was concluded that current efforts of Nigerian public hospitals to provide "safe" blood and CTT fell short of best practices. Provision of apheresis machines, improvement of voluntary non-remunerated donor drive, screening for red cell antigens and antibodies, and availability of iron chelators would significantly improve SCD care in Nigeria. Provision of "safe" blood depends largely on the quality of blood obtained from the donors, who are screened for common viral infections such as hepatitis B, C and human immunodeficiency virus (HIV) and syphilis. The records of donors were reviewed between 1993 and 2000 to determine the prevalence of HIV infection among them and assess their awareness of and attitude towards the virus (Durosinmi *et al*, 2009). Of th

16,080 units of blood collected during the period, 1073(6.7%) were from voluntary donors. The cumulative HIV seroprevalence was 2.1% among commercial donors and 0.3% among voluntary donors. Majority (65%) of the donors were aware of HIV and the role of blood transfusion in the transmission of the virus. Poverty and unemployment among commercial donors were observed to be the main reasons for selling their blood. It was concluded that there is greater risk of transmitting HIV (and possibly other transfusion transmissible infections) through commercial donors.

ARE YOU A VOLUNTARY BLOOD DONOR?
DONORS SAVE LIVES

Automated Exchange Blood Transfusion at OAUTHC, Ile-Ife

Arogundade *et al.* (2014) discussed the challenges of setting up a new therapeutic apheresis service in a resource limited setting, with particular emphasis on the knowledge and use of therapeutic plasma exchange among nephrologists in Nigeria. Therapeutic apheresis (TA) is an extracorporeal blood treatment modality that involves therapeutic plasma exchange (TPE), automated red cell exchange, collection of blood products and haemopoietic cells for transplantation. These treatment modalities are not readily available or affordable being newly introduced in many parts of Africa in the last decade or so. One of such apheresis machines (Figure 21) was acquired by the Department of Haematology and Blood Transfusion to provide automated red cell exchange for patients with SCD who need chronic transfusion therapy. In addition single donor platelet collection can be life saving for patients with severe thrombocytopenia from any cause.

The cost of the procedure is a major limiting factor, but it reduces the prevalence of iron overload in SCD and alloimmunisation and blood transfusion reactions from the transfusion of multiple single units of platelet concentrates.

Blood transfusion reactions were studied at OAUTHC, Ile-Ife, where 462 transfusion were evaluated (Arewa OP, **Akinola NO**, Salawu L, 2009). It was observed that the overall incidence of blood transfusion reactions was 8.7% with febrile non-haemolytic transfusion reaction (FNHTR) being the most common (65%; Table 23). A positive history of previous blood transfusion increased the risk of adverse reactions to blood transfusion (P = 0.0039; Table 24).

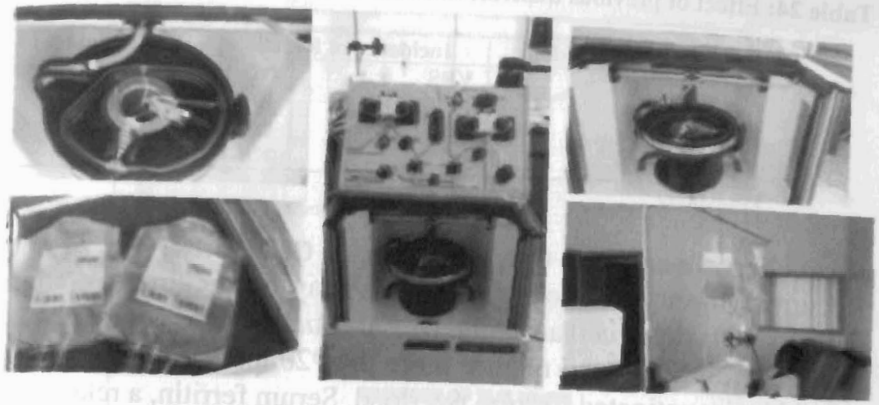

Figure 21: Components of a Therapeutic Apheresis Machine (Cobe Spectra) and a Single Donor Platelet Collection in progress at the Department of Haematology and Blood Transfusion

A recommendation was made to development a national haemovigilance database through the Regional Blood Transfusion Services in the six geopolitical zones in Nigeria.

"NO BLOOD NO LIFE"

Table 23: Incidence of types of blood transfusion reactions seen by ward

Type of Reaction	Number of Blood Transfusion Reactions by Ward (%)				
	Children's Ward	Medical Ward	Surgical Ward	Obstetrics & Gynae Wards	Total (%)
Allergic reaction alone	2 (5)	3 (7.5)	-	1 (2.5)	6 (15)
Febrile non-haemolytic transfusion reaction (FNHTR)	12 (30)	5 (12.5)	6 (15)	3 (7.5)	26 (65)
FNHTR and allergic reaction	2 (5)	2 (5)	1 (2.5)	2 (5)	7 (17.5)
Haemolytic reaction	1 (2.5)	-	-	1 (2.5)	2 (5)
TOTAL	**17 (42.5)**	**10 (22.5)**	**7 (17.5)**	**7 (17.5)**	**40 (100)**

P = 0.58 (no significant difference)

Table 24: Effect of previous transfusion on the incidence of adverse reactions

Previous Transfusion	Incidence of Reaction (%)		Total (%)
	YES	NO	
YES	15 (21.3)	40 (73)	55
NO	23 (11.3)	180(88.7)	203
TOTAL	38	220	258

P = 0.0039

Serum Ferritin and Iron Overload in Sickle Cell Disease

Recurrent top-up red cell transfusions may be required by adult patients with sickle cell anaemia. Drasar *et al.*, (2012) suggested that patients with SCA who require more than 20 units of packed cells should be investigated for iron overload. Serum ferritin, a relatively reliable measure of stored iron (especially when above 1000 µg/l) in the absence of liver iron concentration (LIC), was studied in adult patients with SCA in the steady state and during vaso-occlusive crisis to determine the association between the total number of top-up transfusions and iron status. Fifty two patients comprising 30 in steady state and 22 with vaso-occlusive crisis; and 17 apparently healthy Hb AA controls were investigated (**Akinola** *et al.*, 2014). The mean serum ferritin levels during crisis (919.8±433.3 µg/l) was significantly higher ($p = 0.0103$) than that of the steady state (586.5±457.8 µg/l), which likewise was significantly higher than that of control values (96.0 ±65.5 µg/l; $p < 0.0001$). Seven (23.3%) of the patients in steady state had levels above 1000µg/l and four of them had received 0-7 units of red cell transfusions. The levels of serum ferritin correlated only with the number of total top-up transfusions in this series (Figure 22: $r = 0.517$; $p < 0.01$). It was therefore concluded that serum ferritin was elevated in more than half of the patients in steady state, therefore, it should be routinely measured in patients in steady state and those with levels above 1000 µg/l should be investigated for iron overload irrespective of the number of top-up transfusions received. Serum ferritin concentration in vaso-occlusive crisis was variable and not a reliable assessment of iron status because it is an acute phase reactant.

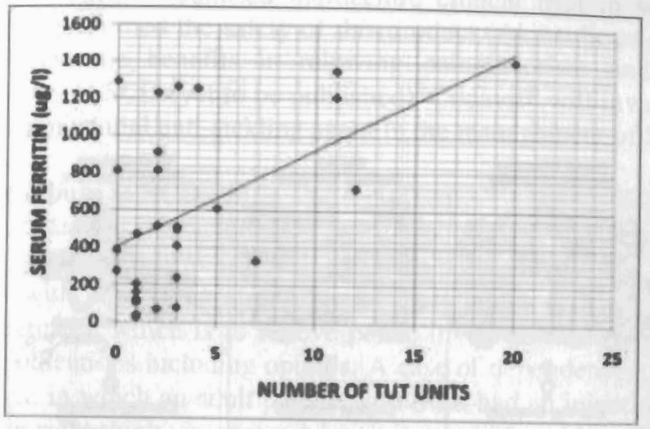

Figure 22: Correlation between Serum Ferritin and the Number of Top-Up Transfusions in the 30 Patients with Sickle Cell Anaemia in Steady State

Disease Modifiers

Since sickling of red cells is central to the development of painful crisis and the complications of SCD, scientists have been working on producing an antisickling agents that will modify the expression of the disease and that will be well tolerated with little or no side effects.

Hydroxycarbamide is the most commonly used disease modifier for SCD. It inhibits DNA synthesis, switches on the production of Hb F, which increased to a mean of 8.6% vs 4.7% in the placebo group (Charache *et al*, 1995), prolongs survival of sickle red cells (by reducing haemolytic rate and increasing the bioavailability of nitric oxide), reduces steady state leucocyte count, and lowers the number of adhesive red cell receptors (Figure 23). Hydroxycarbamide has been recommended for use from early childhood in order to prevent irreversible end organ damage (Fields and Nathan, 2014), but some patients do not respond adequately and may develop severe complications at a young age.

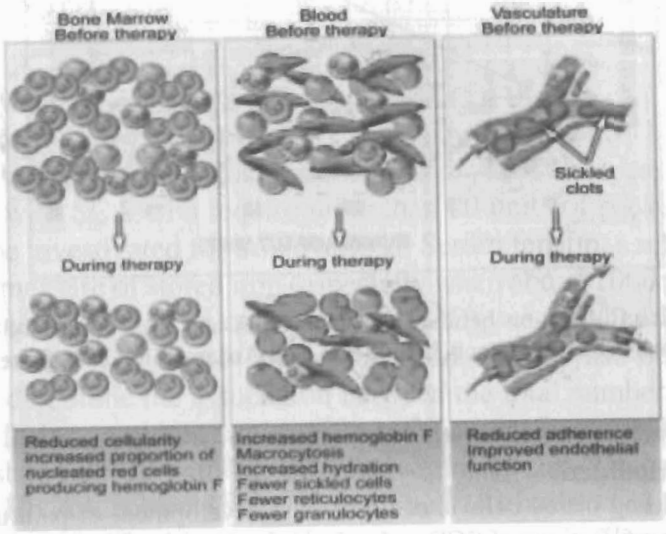

Figure 23: This is a schematic representation of the effect of hydroxycarbamide on sickle cells (Sickle Cell Anaemia – Medscape)

Long term usage may be associated with secondary leukaemia (9.8% vs 3.7%; p>0.05), but the rate is not significantly higher than that of the general population. Other disease modifier s that have been tried are Clotrimazole and Magnesium (both prevent dehydration of sickle cells); and nitric oxide (complicated by the formation of methaemoglobinaemia). Decitabine and 5-azacytidine modify DNA hypomethylation and globin gene expression, but none of these modifiers cure the disease and they have side effects and complications.

Local Herbal Medications

Many herbal remedies and nutritional supplements are available in Nigeria that are being used to prevent painful crisis, some have NAFDAC number and some do not, but randomised multicentre clinical trials have not been done on most of them to justify their use in our clinics. One of such local remedies that has undergone randomised multicentre clinical trial in Nigeria is Ciklavit®, that is derived from *Cajanus cajan*, an edible bean. A

recently concluded randomised multicentre clinical trial in which we participated, confirmed the safety of this product (Akinsulie *et al*, 2009) and confirmed the benefits in relieving painful crises and various manifestations of SCD (yet to be published). Ciklavit® will invariably be an important potential anti-sickling agent in the management of SCD.

Substance Abuse in SCD

Sickle cell disease is characterized by recurrent lifelong episodes of pains of varying severity and frequency in different parts of the body, it is also associated with equally painful severe complications. The mainstay of current treatment, which is to relieve pains, involves the use of a wide range of medications including opioids. A case of dependence syndrome was reported in which an adult patient with SCA had an injection needle stuck in the right thigh, an area used to inject opiods, with an imminent feeling of death. The needle was removed surgically. This case report advocated the need for a multidisciplinary approach to the management of SCA and emphasised the need for psychosocial interventions in adults (Aghanwa *et al*., 1997). Since then, case reports of many pitfalls especially addiction to Pentazocine have been documented among Nigerian patients (Mabayoje *et al*; 2015; Armiya'u A.Y. *et al;* 2016). At OAUTHC, Ile-Ife, a case review over a five year period, of nine individuals with sickle cell anaemia who became dependent on opioids was conducted to highlight some common associated risk factors (**Akinola** *et al*., 2016). The age range of all individuals was between 20-49 years of which 33% were between 20-29 years age group while 55.6% were 30-39 years age group as of last clinic visit. Majority (77.8%) of the patients were females with a male to female ratio of 1: 3.8 and 55.6% had tertiary education as the highest educational level attained. Most (88.9%) were single, while 22.1% were separated from spouse. Majority of the patients (77.8%) were however in an intimate relationship as of the time of this review. Majority 78% were employed and in addition, 88.9% received financial and social support from family members. A total of 22.2% were from homes where the parents were separated. The main drug used in all nine patients was Pentazocine (88.9%), with all having first contact of use introduced by health care professionals while further source of drugs were from pharmacy shops (71.4%). The main route of administration was intramuscular (66.7%), which results in abscess and scar formation (Figure 24). The other common route used is intravenous. There was no increase in terms of poor interpersonal relationship with friends (50%) and siblings (22.2%). Sequel to the persistent use of Pentazocine, these patients finally developed Pentazocine dependency syndrome, 71.4%

were admitted into the mental health ward for treatment, while all (100%) had other associated physical complications. However none of them became abstinent, while two died within the five year review. These preliminary findings suggest that individuals with sickle cell anaemia who become dependent on opioids do not fit the typical profile of uneducated, unemployed individuals with poor social support, instead most of them were highly educated individuals as reported by other researchers (Iheanacho *et al.*, 2015). These findings suggest that iatrogenic introduction and continued self-medication of Pentazocine by individuals with sickle cell anemia probably sustained the habits which ultimately led to dependence, as previously observed by Mabayoje *et al.*, 2015; Kotila *et al.*, 2015; Armiya'u A.Y. *et al.*, 2016.

It was concluded that there is a need to control the use, availability and regulation of Pentazocine in the management of pain in individuals with sickle cell disease. Physicians were cautioned in respect of the use of Pentazocine as a first line routine medication to relieve pain in patients with SCD.

Figure 24: Injection abscess and scars on the right thigh of an adult with SCA

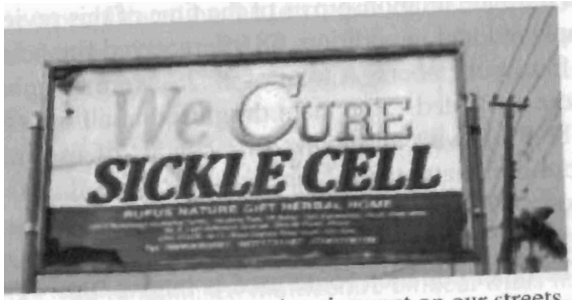

Figure 25: A common advertisement on our streets

Haematopoietic Cell Transplantation

Haematopoietic cell transplantation (HCT) is the only available cure for SCD since gene therapy and genome editing are still undergoing clinical trials. These modalities of treatment are addressing the root cause of the disease. The first patient with SCD to have bone marrow transplantation (BMT) had Hb SS and acute leukaemia (Johnson *et al*, 1984) with a good outcome. After this first report, two groups in the USA (Walters *et al.*, 1996) and Belgium (Vermylen *et al.*, 1993) published the results of the first series of patients with SCD that had allogeneic BMT, with overall survival outcomes >90% and event-free survival > 85%. Other sources of haemopoietic stem cells (HSC) include peripheral blood, cord blood and foetal liver. Haemopoietic cell transplantation (HCT) in SCD involves harvesting adequate quantities of haemopoietic stem cells (HSC), which are the CD34+ CD 38- cells and transfusing them into the immune incompetent recipient; the use of reduced intensity bone marrow ablation using busulphan and cyclophosphamide to reduce the immune response of the recipient to prevent graft versus host disease (GvHD); and manage complications that may occur from the immune incompetent state. Early procedure related mortality occurs in 5-10% of cases; cure rates are between 71 and 82%; and graft versus host disease (GvHD) occurs in < 15% (incidence is lower with umbilical cord stem cells). The recommended criteria for BMT eligibility are age less than 16 years; low baseline Hb F; high baseline leucocyte count (>15 x 10^9/l); and one or more of the following: renal insufficiency, acute chest syndrome, red cell allo-immunisation (> 2 antibodies) or failure of iron chelation while on chronic transfusion therapy. Long term BMT complications include infertility and acute leukaemia. Adults preferably less than 40 years old are now receiving HCT with good outcomes.

In preparation for the haemopoietic cell transplantation (HCT) service proposed to OAUTHC, Ile-Ife, in 2008, the yield of HSC was investigated in peripheral blood of adults (without mobilization) and cord blood (Aloko 2018). In an attempt to obtain the haematologic indicators that may be useful in selecting prospective donors that would produce a good harvest, the correlates of CD34+ cell count were determined (Aloko *et al.*, 2018) in the peripheral blood of 50 adults and cord blood 50 newborns. It was observed that the mean CD34+ cell count in cord blood samples (8.71 ± 8.87/μL) was significantly higher than that of adult blood (1.90 ± 1.43/μL; P = 0.0001; Figure 26). This confirmed the report of Perumbeti *et al* (2012).

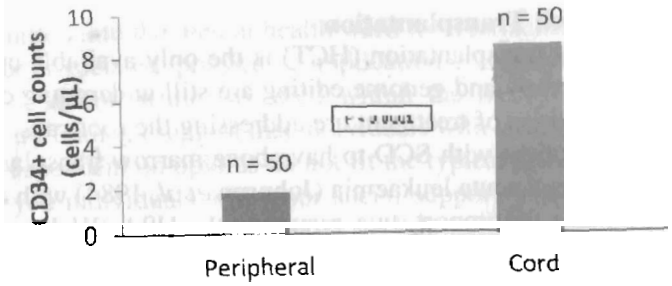

Figure 26: CD34+ Cell Count in Peripheral Blood and Cord Blood

Individuals with sickle cell trait (Hb AS, 18%) had more CD34+ cells (2.92 ± 1.75/μL) in circulation than those with Hb A only (1.68 ± 1.27/μL; P = 0.016; Figure 27). Cord blood of newborns from mothers with Hb AS (32%) had a significantly higher CD34+ cell counts (12.32 ± 11.27/μL) than those with Hb A only (7.01 ± 7.05/μL; P = 0.047).

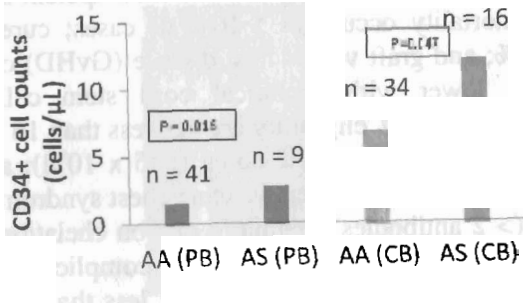

Figure 27: The CD34+ cell count in peripheral and cord blood and haemoglobin types

The haemoglobin type of mothers in this study may therefore predict the yield of CD34+ cells that may be harvested from the cord blood ($P = 0.047$) of their babies irrespective of the Hb type of the babies. According to Martin-Antonio *et al.* (2011) donors with haemoglobin type A only had the lowest yield of CD34+ cells. A weak positive correlation was obtained between CD34+ cell counts and PCV in cord blood in the present study (r = 0.335; P = 0.017; Figure 28). This result was contrary to that of Mehta *et al* (2001). Also, the CD34+ cell counts showed a weak positive correlation with Hb concentration (Figure 29), but the strength of these

correlations in cord blood were however not sufficient to make PCV and Hb conc surrogate markers of CD34+ cell counts in newborns.

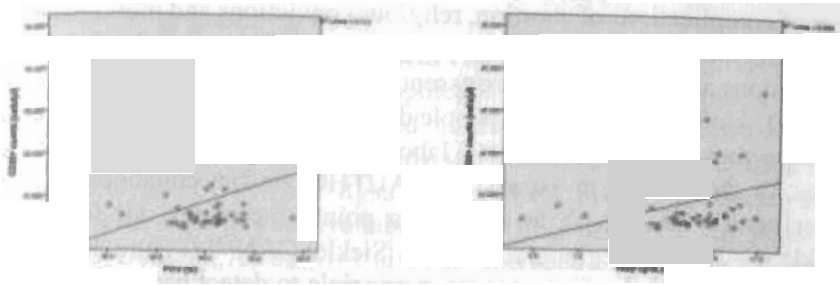

Figure 28: Correlation between CD34+ Cell Count and Packed Cell Volume in Cord Blood (r = 0.335; P = 0.017)

Figure 29: Correlation between CD34+ cell count and haemoglobin concentration in cord blood (r = 0.300; P = 0.034)

This study is the first to document that cord blood is a better source of HSC (CD34+) cells than adult blood per unit volume in apparently healthy non-mobilized Nigerians and confirm reports from elsewhere. Adults with SCT and newborns of mothers with SCT (Hb AS) were more likely to have higher CD34+ cell counts than those with Hb A only. There was a tendency for CD34+ cells to increase as haemoglobin concentration and haematocrit of cord blood increased, but CD34+ cells should be counted to assess the yield in a HSC harvest before being transplanted as no surrogate haematologic indicator was identified.

"The Life of the Flesh is in the Blood; No Blood No Life"

Preventive Strategies - Primary Prevention

Prevention is better than cure. Primary prevention is it. It involves counselling and testing for the sickle gene and thus preventing the transfer of the gene to the next generation. This can be done if one is a carrier of the Hb S gene by refusing to marry another carrier or marry and refuse to have children or have children that will be investigated for the disease prenatally. To this end a multicentre survey of the acceptability of prenatal diagnosis of SCA by female patients with SCA and parents of individuals with SCA (Durosinmi *et al.*, 1995); and a sample of the Nigerian population (Durosinmi *et al.*, 1997). These surveys showed that

psychosocially, female SCA patients, their parents and the general public accepted the use of prenatal diagnosis (PND) as a means of preventing SCA. However, fewer patients would opt for termination of pregnancy for fear of complications of abortion, religious convictions and moral reasons. Knowing one's "genotype" and having the ability to make informed discussions after counselling are essential for effective primary prevention of SCD. Unfortunately, many people do not know their haemoglobin type because they lack the awareness, laboratories are too far away in urban areas, or the test is too expensive. OAUTHC, Ile-Ife, collaborated with other centres in Nigeria to evaluate a point of care test kit for SCD, SickleSCAN™ (Nwegbu *et al.*, 2017). SickleSCAN™ (Figure 30) uses a qualitative lateral flow immunoassay principle to detect haemoglobins A, S and C. It is a screening test for SCD with a diagnostic sensitivity, specificity and test efficiency for SCD (Hb SS and Hb SC) of 100.0, 98.2 and 98.2%, respectively when compared to HPLC. The prevalence of SCD was observed to be 3.4%. This study showed SickleSCAN™ to be a viable screening tool that can easily be used in primary health care setting or community-based studies for early diagnosis of SCD with little expertise and low cost, thus ideal for this environment.

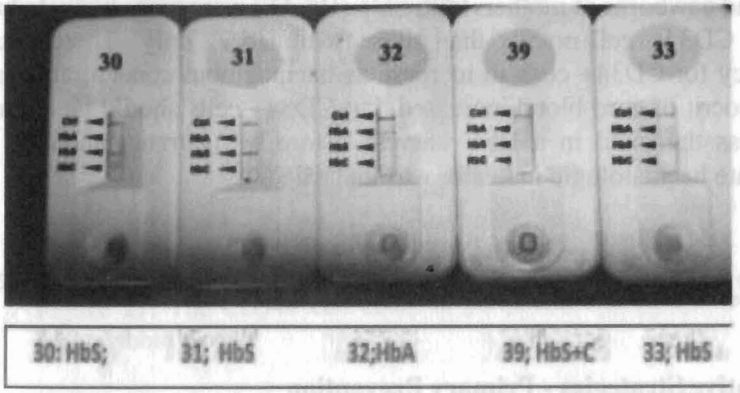

Figure 30: Samples of cassettes from SickleSCAN™ screening tests showing the separation of the various haemoglobins (Nwegbu *et al.*, 2017)

In addition, newborn screening conducted with the use of HPLC or IEF enables early detection and diagnosis of new cases who should be followed up and managed strictly to prevent complications. Adelasoye (2013) screened 380 newborns and observed that the incidence of Hb S only was 1.8%, Hb A only 82.4%, Hb AS 13.4% , Hb AC 1.6%, and Hb SC 0.3%

(Figure 29). The mean Hb F in newborns was 65.6 ± 8.6%. This study concluded that the incidence of Hb AS and Hb SS in newborns at OAUTHC is less than previously reported in Nigeria.

Secondary Prevention – Use of Local Herbs as Antisickling Agents

Any intervention outside haematopoietic cell transplantation or gene therapy/genome editing is regarded as attending to the down-stream effects of the disease. The use of hydroxycarbamide has revolutionized the management of SCD, but it is not without its side effects and complications. Also it is not available to all in need because it is expensive to sustain. Obafemi Awolowo University included Sickle Cell Research as part of her strategic plan (2010-2015) with a view to engaging in research that will contribute substantially and innovatively to the national economy and the well-being of the patients. The Sickle Cell Project for the Prevention and Management of SCD was funded by TETFUND between 2012 and 2015. This project enabled the Drug, Research and Production Unit (DRPU) and Department of Haematology and Immunology to research into the many local edible herbs such as *Telfairia occidentalis* (Cryil-Olutayo *et al* 2018a), *Moringa oleifera* (Cryil-Olutayo *et al* 2018b) and *Cnidoscolus aconitifolius* (Cyril-Olutayo, 2015) that have antisickling properties and effects with no significant side effects. The leaf extract of *Cnidoscolus aconitifolius* (mill.) I.M. Johnst (identified and authenticated by G. Ibhanesebor at the IFE Herbarium, OAU, Ile-Ife with voucher number IFE 17256) amongst others was investigated to determine the mechanism of action of its antisickling properties. *Cnidoscolus aconitifolius* (CA) is an edible vegetable rich in nutrients, vitamins, proteins, antioxidants and is being used locally in the management of sickle cell anaemia. This plant is known as "Efo Iyana Ipaja", "Efo Jerusalem", in southwestern Nigeria (Awoyinka *et al*, 2007), while in the Niger Delta of Nigeria, it is called "Hosiptal Too Far", because of the numerous traditional claims that it improves the blood. The antisickling properties *in vitro* are probably due to the effect of the plant extract to increase the mean cell volume (MCV) in each fraction, particularly in F3 and F4, significantly ($p < 0.05$) after treatment with CA extract, when compared to untreated cells, but this was not so with Ciklavit®. The plant extract also reduced the percentage of dense cells after incubation (Figure 31; Table 23) and prevented a significant reduction in the membrane stability (Figure 32) when compared to a positive control (Ibuprofen). This study authenticated the use of this plant for use as an antisickling agent.

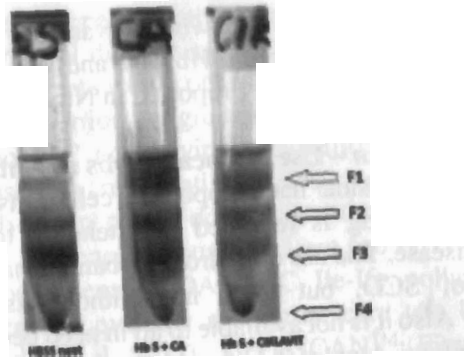

Figure 31: Discontinuous density gradient of sickle cells before and after incubation with plant extract

Table 23: Fractions (F2, F3 and F4) from discontinuous density gradient (n=3; *values are presented as mean ± SEM)*

Fractions	Mean RBC of	Mean RBC of Hb	Mean RBC of Hb
F2	5.49	5	21
F3	34.33 ± 3.40	55.67 ± 5.87	48.43 ± 1.66
F4	46.01 ± 2.11	19.56 ± 6.54	29.98 ± 1.69

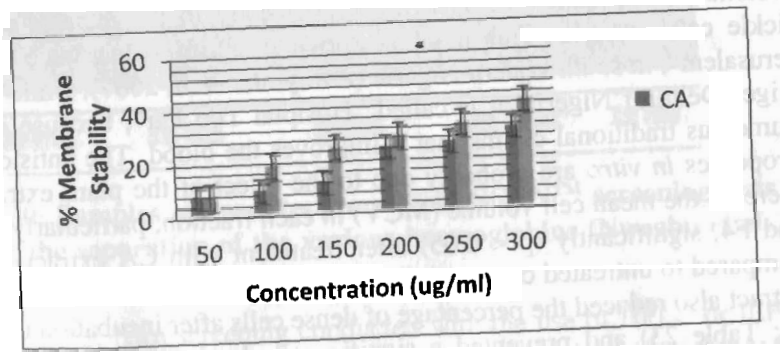

Figure 32: Dose related membrane stability effect of CA extract on sickle cells (Hb SS)

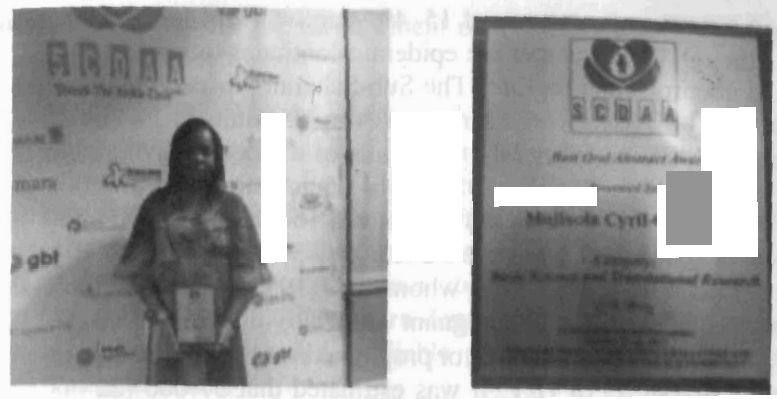

Figure 33: My first PhD graduate won the "Best Oral Abstract Award" at SCDAA Convention, Atlanta Georgia, October, 2017

The possible mechanisms of action and its antisickling effects *in vitro* compared to Ciklavit® were presented at the 45th convention of the Sickle Cell Disease Association of America (SCDAA) at Altanta, Georgia October 25-28, 2017 where it won the best oral abstract award (Figure 33; Cyril-Olutayo *et al* 2017). Clinical trials are being awaited.

HUMAN IMMUNODEFICIENCY VIRUS (HIV) AND ACQUIRED IMMUNE DEFICIENCY SYNDROME (AIDS)

This lecture would not be complete if I do not talk about human immunodeficiency virus (HIV), a retrovirus that causes chronic infection in man. It infects immune cells that are CD4 positive (lymphocytes, macrophages, dendritic cells) through which it gains access, multiplies rapidly and destroys the cells. When the immune cell (CD4) count is so low, that is, less than 200 cells/cmm, clinical features of acquire immune deficiency syndrome (AIDS) occur, such features include, weight loss (>10% body weight within a month), fever and diarrhoea that persist for more than one month. World AIDS Day is celebrated on the first day of December every year to remind people that AIDS is real, although the HIV infection may not show on the face initially. Since the beginning of the epidemic, WHO reported that more than 70 million people have been infected with the HIV virus and about 35 million people have died of AIDS. Globally, 36.9 million (31.1–43.9 million) people were living with HIV at the end of 2017. An estimated

0.8% (0.6-0.9%) of adults aged 15–49 years worldwide are living with HIV, although the burden of the epidemic continues to vary considerably between countries and regions. The Sub-Saharan African region remains most severely affected, with nearly 1 in every 25 adults (4.1%) living with HIV and accounting for nearly two-thirds of the people living with HIV worldwide. In Nigeria, UNAIDS (2016) documented that 220 000 (150 000 - 310 000) had new HIV infections and 160 000 (110 000 - 230 000) AIDS-related deaths. There were 3 200 000 (2 300 000 - 4 300 000) people living with HIV in 2016, among whom 30% (19% - 42%) were accessing antiretroviral therapy. Of the pregnant women living with HIV, 32% (22% - 44%) were accessing treatment or prophylaxis to prevent mother-to-child transmission (MCT) of HIV. It was estimated that 37 000 (22 000 - 56 000) children were newly infected with HIV due to MCT. Among people living with HIV, approximately 24% (18% - 32%) had suppressed viral loads (VL). Osun State is still one of the many states with low prevalence of HIV. The mode of transmission is through the blood-blood contact. The risk factors are unprotected heterosexual intercourse; mother to child transmission; blood transfusion; needle stick injuries and sharing of sharp objects (IDU), surgery etc.; homosexual interactions. Human immunodeficiency virus cannot be contracted from kissing, handing hands, sharing the same crockery or toilet seats, but prevention is better than cure. The **ABC** of prevention are **A**bstinence, **B**e faithful or use the **C**ondon and if these cannot practised then the likelihood of acquiring the infection is high. Then **DEF** measures should be taken, i.e. **D**iagnosis, **D**rugs (HAART) or **D**eath, **E**ducation (on living positively), **F**aithfulness in or **F**ailure to take medications will lead to VL undetectable or insuppressible VL due to the development of resistant strains, respectively. Just like SCD, there is no medical cure for HIV infection, therefore medications must be taken for life in the right combination, at the right time. Only 100% adherence can prevent resistant strains from developing.

Before international agencies came to assist Nigeria deal with the AIDS epidemic, highly active antiretroviral (HAART) agents were expensive and not accessible, so Prof. Femi Soyinka (retired) collaborated with a team at the Centre for Special Studies, New York Presbyterian Hospital, NY, USA, under the Starfish Project. This project provided medications for the treatment of some of our patients then, so there was a need to determine who should commence (HAART). The World Health Organisation (WHO) recommended, at the time, the use of total lymphocyte count (TLC) as a substitute for CD4 count, which was not

readily available, for the management of individuals living with HIV in resource-limited settings.

Akinola *et al* (2004) analysed the TLCs and CD4 counts of 109 patients attending the clinic at OAUTHC, Ile-Ife and observed that using the cut-off value of <1,200/cmm for TLC to commence treatment, 37.8% of the patients who had TLC >1,200/cmm had CD4 counts less than 200 which was the recommended value for commencing HAART in resource-unlimited settings. It was therefore concluded that since one in three patients with HIV would have been deprived of treatment using the TLC cut-off value, TLC was not a reliable predictor of CD4 count in HIV-infected individuals.

Since then things have changed significantly and WHO is now recommending a "test and treat", protocol for the management of HIV infection with a goal to identify 90% of people living with the virus; treat 90% with HAART; and get 90% of those on HAART to viral load undetectable status by 2020. **This is the vision 90: 90: 90 by 2020.**

"NO BLOOD NO LIFE"

AWARDS, FELLOWSHIPS AND GRANTS

I received a Wellcome Trust Fund Award (UK) to study the Rheology of Sickle Cell Anaemia for my PhD (1990-1992).

I was elected a member of the American College of Physicians in 2001 and was later nominated by the then President of the West African College of Physicians, Dr. Sonny Kuku, for a three-month International Exchange Fellowship Program for the study of HIV/AIDS in America between April and July 2004, at the Centre for Special Studies, New York Presbyterian Hospital, NW, USA, supported by Pfizer Int. Plc. The experience I obtained was novel and I was requested to present a paper titled, ***"The Brain Exchange: Insights Exported and Imported via ACP's International Fellowship Exchange and International Speakers' Program International Committee"***, at the next scientific conference and annual meeting of ACP in San Francisco, USA, April 2005, I was given the award of Fellow of the American College of Physicians.

NECAIN Project Award for the Care and Support of PLHIV in Osun State, sponsored by USAID (2008-2010) was given to Mustard Seed Health Awareness Initiative, Ile-Ife, under my watch.

I have been a beneficiary of many other local, national and international research grants including:

OAUTHC grant for the study of some observations in the cardiovascular status of Nigerian individuals with sickle cell anaemia at rest and in response to exercise (1992);
OAU grant for the study of CD4/CD8 in health and disease (2004);

Niemeth International Pharmaceuticals Plc grant for a Multicentre Clinical Trial on the Ciklavit® and SCA. (2004-8);

OAU research grant to study the cardiovascular risk and lipid profile in HIV infected individuals on ARVs. (2005);

Glaxo-SmithKline Multicentre Clinical Trial on the advantage of chlorproguanil-dapsone-artesunate (CDA) over chlorproguanil/dapsone (LAPDAP) in the treatment of uncomplicated *F. falciparum* malaria infection. (2005-7);

Federal Ministry of Science and Technology Clinical Trial on the antiretroviral effect of Garsuma. (2007-8);

OAU TETFUND research grant to develop and produce a herbal drug for the management of sickle cell disorder and advocacy activities. (2012-2015).

National Research Fund (NRF) – TETFUND 2018 to study the molecular and immunohistochemical characterization of some cancers in southwestern Nigeria: the role of infections and human leucocyte antigens

MY CONTRIBUTIONS TO TRAINING IN HAEMATOLOGY AND INTERNAL MEDICINE IN NIGERIA

Since I joined the Department of Haematology and Immunology as a Lecturer I/Senior Registrar (Internal Medicine) in 1993, I have served the University in various capacities including Head of Department (2011-13; acted for many years), Chairman, Postgraduate Committee (2002-2007), Chairman, Research Committee (2010-2013), Vice-Dean (2007-2008) and Dean (2013-2015), Faculty of Basic Medical Sciences. As a teacher, my first MSc and PhD students both graduated in 2015. In October 2016, I was invited to examine the first MD candidate in the Department of Medicine, Ahmadu Bello University, Zaria, where I was consequently, invited to assist with the training of postgraduate students in the Department of Haematology as a visiting professor.

I have participated in the care of patients with sickle cell disease, other types of anaemia, haemato-oncological disorders, coagulopathies and HIV/AIDS attending various clinics at Obafemi Awolowo University Teaching Hospitals Complex (OAUTHC), Ile-Ife. I have contributed to

the training of many resident doctors and many supernumerary trainees in various disciplines from other teaching hospitals and medical centres from all over the country that are now consultants and even professors today (Prof. Baba Kagu, Prof. Rasaq Adebayo, Prof. Vincent Mabayoje). I have also supervised over 10 residents in Haematology and Internal Medicine in their Part II dissertations. One of them by, Dr. Rasaq Adebayo, won the SF Kuku Prize for the best overall candidate at the April and October 2001 Final Fellowship Examinations of the Faculty of Internal Medicine of the West African College of Physicians (Figure 34).

I am a resource person for the Revision and Update Courses of both the National and West African Postgraduate Medical Colleges. I am an examiner at that the National Postgraduate Medical College in the Faculties of Pathology and Internal Medicine. I was elected a member of the Board of the Faculty of Pathology at the National Postgraduate Medical College in November 2005 and I was elected the Treasurer of the same Board in November 2007 for a period of two years. In 2016 I was nominated to be a member of the Court of Examiners in the Faculties Pathology and Internal Medicine (Chairman of Haematology). I am a reviewer for many reputable journals nationally and internationally.

Figure 34: Dr. Rasaaq Adebayo, now Professor of Cardiology, receiving the SF Kuku Chairman Prize in Internal Medicine for the best overall candidate in the April and October 2001 Final Fellowship Examinations of the Faculty of Internal Medicine of the West African College of Physicians, Freetown, Sierra Leone.

In 1995, during my first tenure as acting Head of the Department, I was elected a member of the interim executive to resuscitate of the moribund Nigerian Society for Haematology and Blood Transfusion (NSHBT). This interim executive organised a conference in UCH, Ibadan on chronic myeloid leukaemia where I was elected the Assistant Secretary for the Society, a position I occupied consecutively for four years. During that time, in 1997, a very successful International Conference of the Society (25th Scientific Conference/AGM) was organised at OAU, Ile-Ife and I was the Chairman of the LOC. The theme was the Management of Anaemia. In 1999 I was elected Secretary of the NSHBT and re-elected in 2001. While in office I took up the role of Editor for the Newsletter of the Society and facilitated the publications of two editions. In Vol. 4 No. 1 (2000/2001) edition I wrote an editorial titled, ***The New Millennium - What has it to offer the Nigerian Haematologist?*** In Vol. 5 Nos. 1 & 2 (2002/2003) edition the editorial was titled, ***Anaemia: A common Indicator of Disease – What's New.*** Many years later, I was nominated the first Editor-in-Chief of the Nigerian Journal of Haematology (NJH) in 2016 at the 42nd Annual Scientific Conference/AGM in Lagos, Nigeria. To the glory of God, I led the Editorial Board to successfully publish the first edition of the Journal, which is the official journal of the Nigerian Society of Haematology and Blood Transfusion, on 22nd August 2017, at our 43rd Annual Scientific Conference/AGM in Kano. It is the first African journal of haematology published by a professional association/society to disseminate information obtained from research outputs.

In continuance of my service to NSHBT, I was the LOC Chairman for the annual meeting in 2008. The theme of the conference was "Haematopoietic Stem Cell Transplantation – Which way Nigeria?" It was held at the Royal Park Hotel, Iloko-Ijesa and it was well attended by Haematologists from all over Nigeria. The guest speakers were Prof. Axel Zander from the Bone Marrow Transplant Unit, University of Hamburg, Germany and Mrs. Helen Baldemero from the European BMT Registry, Switzerland. At the meeting I presented a paper on the knowledge and attitudes of Nigerians in the university community towards organ/tissue transplantation. Consequently, a proposal for the provision of haematopoietic stem cell transplantation at OAUTHC is on ground and if successfully implemented should be the next centre for Nigeria. Unfortunately again, funding is a major limiting factor. We need the

support of well-meaning Nigerians and collaborators worldwide. The National Institutes of Health, Bethesda, Maryland, USA, has shown interest in collaborating with us to train our staff.

Recently, physicians for children and adults living with sickle cell disease in Nigeria got together to form a Sickle Cell Support Society of Nigeria (SCSSN) for which I was nominated the Regional Coordinator for the South West. Prof. Adekunle Adekile, one of our Paediatricians in Kuwait, is the President of the Society. I also chaired the LOC that planned the first conference of the Society in June 2013 to which Dr. Baba Inusa, a Paediatrician from the Evelina Hospital for Children, London UK, was invited as the Guest Speaker. I also coordinated the training workshop on "Newborn Screening in Nigeria", in collaboration with Biorad, Nigeria, at the conference. The following month I led a team from OAUTHC, Ile-Ife, Nigeria, to University of Benin Teaching Hospital, Benin City, Nigeria for a two week training workshop in Haematopoietic stem cell transplantation. During the workshop our team participated in the transplantation of a sickle cell patient who received stem cells from the bone marrow of a compatible sibling. The outcome was good.

In June 2014, I lead the first red cell exchange transfusion procedure using an apheresis machine, "Cobe Spectra", to treat prolonged priapism, a complication, in an adult male patient with sickle cell disease at OAUTHC, Ile-Ife. In 2016 I was invited to train resident doctors and consultants on the use of the Cobe Spectra apheresis machine at the National Hospital, Abuja.

SERVICE TO THE COMMUNITY

In 1995, 5th December precisely, I was inaugurated the President of the Sickle Club, Ile-Ife by Prof. Akin Akinyanju, President of the Federation of Sickle Cell Clubs in Nigeria (FESCON), who later became the founder of the Sickle Cell Foundation, Lagos, Nigeria. I was re-elected in 1999/2000 for another tenure and remained President until February 2005 when I handed over to the late Dr. Adediran, a consultant Haematologist. The **Sickle Cell Club (SCC) Ile-Ife is a non-governmental organisation** that cares for sickle cell patients and provides information to increase awareness and ultimately prevent the occurrence or reduce the prevalence of the disease in the community. During my tenure, I facilitated many workshops, seminars, symposia and out-reach campaigns (particularly in secondary schools and market places) in some parts of Osun State. Fund-raising activities were conducted from time to time to assist affected

individuals who had financial challenges to settle hospital bills. The Club had the support and blessing of the community and this is evident by the role our late Patron, The Ooni of Ife, Oba Okunade Sijuwade, Olubuse II, played in donating two acres of land, to the Club in 2003 which was unfortunately taken away from us a few years ago after his demise because we lacked funds to develop it beyond the foundation level.

Since 2010, SCD is celebrated on the 19th day of June every year and as part of community aspect of the Sickle Cell Project, OAU in 2012 collaborated with an NGO called Mustard Seed Health Awareness Initiative (MSHAI), Ile-Ife, Osun State, to study the prevention of SCD through advocacy in two states in south-western Nigeria, Osun and Ondo States. In line with the stated objectives of the study 25 schools from 10 LGAs in Osun State and 30 schools from eight (8) LGAs in Ondo State participated in the project. A module for the training of peer educators was developed for SCD and used to train senior secondary school students (SS1) and their teachers. The registry of clients with SCD attending health facilities at all levels in both states developed was an incomplete database with inadequate information. Awareness of SCD was increased in the targeted States through community mobilization and distribution of information, education and communication (IEC) materials. Sickle Cell Clubs and SGs were established and supported in each state.

In 1998 I was elected the President-elect for the Medical Women's Association of Nigeria (MWAN), Osun State Branch, but a year before that, I was nominated to work with other medical women to facilitated the establishment of the Creche at OAUTHC with the support of the Management of the hospital. The Creche was subsidised by the OAUTHC to provide care for children of working mothers while they were at work. The day-to-day running of the creche however was organised by the medical women and the staff we employed. As the President of MWAN (2000-2003), the Association held her first biennial scientific conference and general meeting in February 2002. The theme for that conference was, ***"Adolescent Sexuality"***. It was well attended by students from various Secondary Schools within and around Ile-Ife and many scientific papers were presented. Towards the end of my tenure, MWAN set up a "Well Woman Clinic", at one of the Community Health Centre of OAUTHC, Eleyele, Ile-Ife, with the approval of the CMD. The clinic is still running till today and it has helped to save the lives of some of our women who would have died from one cancer or the other.

In 2005 I anchored a television series on **"You and Your Health"**. This thirty-minute weekly talk show on **NTA-Ife** was produced by Mr. Jide Onifade. I invited specialist doctors from various fields to discuss and educate the people in the community on topical health issues, thus empowering them to improve the quality of their lives. Viewers were allowed to ask questions using a text messaging system (SMS). The programme was watched and enjoyed by many people in Ile-Ife and environs for 26 weeks between September 2005 and April 2006 and then again between November and December 2007. It was supported by some dignitaries in the community.

In January 2008 I was invited by an organisation called NELA Consortium for AIDS Initiative in Nigeria (NECAIN) and received an award, supported by USAID, to coordinate a project for the care and support of people living with HIV (PLHIV) in Osun State in collaboration with Society for Women and AIDS in Africa, Nigeria (SWAAN). To do this project effectively, I had to receive training in palliative care and participate in training many PLHIV and community health extension workers (CHEWs) in home based care for PLHIV. Ultimately, an NGO called the Mustard Seed Support (MSS) Network, which later became registered by CAC as Mustard Seed Health Awareness Initiative (MSHAI) was established. This NGO, under my watch as the Executive Director, has formed many support groups (SGs) in Osun State under the NECAIN Project, in addition to the existing Ife Starfish Support Group (ISSG) established by the team on the Centre for Special Studies, New York Presbyterian Hospital, NY, USA, for PLHIV in OAUTHC, Ile-Ife.

Mustard Seed Health Awareness Initiative supports people living with different types of cancer and a support group for people with chronic lymphocytic leukaemia has recently been formed in Ile-Ife, in collaboration with **Chronic Lymphocytic Leukaemia Advocate Network (CLLAN), UK**. Mustard Seed HAI provides the enabling environment for people living with chronic illnesses to meet without fear of stigmatization or discrimination and it networks with other NGOs and institutions to improve the quality of life of the clients and facilitate access to income generating schemes in Osun State for the needy.

CONCLUSION AND RECOMMENDATIONS

Mr. Vice-Chancellor, Sir, having presented some aspects of my research interest in an hour, I hereby conclude using the framework of the argument that says, "the management of sickle cell disease should be targeted at the root cause of the matter, rather than to manage the down-stream effects of the disease". The root cause of SCD is the point mutation that produces sickle blood cells, which can be cured by haematopoietic cell transplantation or gene therapy/genome editing. These modalities of treatment are not readily available in this part of the world on the one hand and on the other hand, they are expensive and yet not without complications that may be fatal. If, however, they are available, then every individual with SCD should be given the opportunity to obtain cure. In the meantime, we must all endeavour to join hands together to prevent the continuous transfer of the gene to future generations yet unborn and manage the down-stream effects in those who are presently living with the disease to prevent complications and thereby prolong lives through best practices in our clinics and in our homes. The family support system in place in Nigeria has helped to prolong the lives of many of our patients with SCD. My little sister that was born in London in 1970 was given five years to live, but when she was brought to Nigeria at the age of four years her life was prolonged by eight years before she succumbed at the age of 12 years to severe anaemia probably following malaria infection and sequestration crisis. She might still be alive today if facilities were available for emergency exchange blood transfusion. Many people have lost their lives to this disease and more will if the government of Nigeria does nothing about it. I therefore recommend that facilities for emergency exchange blood transfusion be made available in every tertiary hospital (at least one per state to start with) and all individuals with SCD be enrolled under National Health Insurance Scheme (NHIS) that will make available hydroxycarbamide and other medications needed to maintain the steady state for as long as possible without side effects. The day when SCD will be curable for all by managing the root cause with genome editing is near, but in the meantime, let us all join hands together to prevent the inheritance of SCD and manage the down-stream effects in the best way possible.

"NO BLOOD NO LIFE"

FUTURE PLANS

There are still many areas to study in individuals with SCD in Nigeria and in haematology at large, but this can only be done one by one depending on availability of funds.

We have recently completed a study to further understand iron status in SCD in relation to the genes that regulate iron levels in the body. The data shall be published soon.

The OAU Sickle Cell Project under the leadership of Prof. Joseph Agbedahunsi (Drug Research and Production Unit, Faculty of Pharmacy) is still in the process of formulating a herbal medication for the management of the down-stream effects of SCD. It is hoped that a clinical trial will soon be embarked upon that I will coordinate as soon as funding is available to confirm the *in-vitro* observations made so far.

Plans are on the way to establish a haematopoeitic cell transplant programme at OAUTHC, Ile-Ife, in collaboration The Sickle Cell Unit of the National Institutes of Health, Bethesda, Maryland, USA, under the leadership of Prof. Swee Lay Thein and Prof. John Tisdale. Prof. Victor Adetiloye, CMD, OAUTHC, Ile-Ife, and his Board members are committed to this programme. It is hoped that a stem cell laboratory and a haematopoietic cell transplant unit will be put in place soon to make this vision achievable and I pray that you will all be a part of this success story one way or the other.

In addition, there are plans to characterise certain common cancers in south-western Nigeria under the leadership of Prof. Kayode Adelusola and myself in collaboration with Prof. Funmi Olopade, University of Chicago, USA and others.

"The Life of the Flesh is in the Blood; No Blood No Life"

CLOSING REFLECTIONS

Alas, Mr. Vice-Chancellor, Sir, I have come, I have seen, but I have not yet fully conquered. "Ni nu o fii o laa ni omo pandoro mi gbo." When I returned to the shores of Nigeria with my PhD in 1992, thought I would be able to achieve my vision given 20 years or so, but through thick and thin I am nearer my vision today than ever before. It has been a longer journey than I envisaged, but above all I did not give up, because it is "not

by might, nor by power, but by my spirit saith the Lord of hosts" (Zechariah 4: 6; KJV). God Almighty surrounded me with people who contributed positively or negatively to my success as a teacher, a researcher or as a service provider. So, invariably Team work has contributed significantly to my being able to stand before you today, for **T**ogether **E**veryone **A**chieves **M**ore, therefore, the multidisciplinary approach is the best approach to providing standard-care practices and improving the quality of life of our patients. I would like to admonish those coming after me to remain focused on their vision, not to give up and remember that "a tree does not make a forest"; the more the merrier. They must try to enjoy their work no matter their circumstances, because, "all things work together for good to them that love God, to them who are called according to His purpose", (Romans 8: 28; KJV).

At this point, Mr. Vice-Chancellor Sir, distinguished ladies and gentlemen, permit me to express my appreciation to my family at home and abroad, especially my darling husband Prof. David Olayinka Akinola, former CMD of OAUTHC, Ile-Ife (1997-2005), my son, Dr. Ikeoluwapo **Akinola**; my brothers, the Bamgbaiyes; and our late parents for contributing to the success of my career by supporting me in various ways throughout my academic pursuit. I am extremely grateful. To my mentors, motivators and trainers, especially Prof. Muheez Durosinmi, Prof. Ibironke Akinsete, Prof. Akinyanju, Prof. Aba Sagoe, Prof. Michael Balogun, Prof. Lucio Luzzatto, Prof. Ghullam Mufti, Prof. John Stuart, Prof. Gerard Nash and the late Prof. John Goldman; my colleagues, residents, friends and foes, students, patients and sponsors/donors; and members of the Department of Haematology and Immunology at OAU and OAUTHC, Ile-Ife. I say a big thank you for giving me the opportunity to work with you on my journey to acquiring knowledge and impacting the community.

"BLOOD IS LIFE"

To God be the glory, great things He has done. Thank you for your attention. God bless you all.

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