CURRENT CONCEPTS IN THE MANAGEMENT OF SICKLE CELL ANAEMIA

Korubo K.I, Nwauche C.A.N
Department of Haematology & Blood Transfusion, University of Port Harcourt Teaching Hospital, Rivers, Nigeria.

Correspondence:
Korubo K.I., Consultant Haematologist, University of Port Harcourt Teaching Hospital, Rivers State, Nigeria. dadakorubo@yahoo.com
Tag Words: Sickle Cell Anaemia, recent management, Nigeria

ABSTRACT
Background: *Sickle cell anaemia is the commonest genetic disease in the world and is caused by a mutation in the β-globin chain gene of the haemoglobin. Consequently the variant haemoglobin formed easily polymerizes under hypoxic conditions leading to the clinical manifestation of the disease which is highly variable. SCA is associated with a high morbidity and mortality rate especially in children less than five years in Sub-Saharan Africa where the disease burden is highest geographically.*
Aim: *To review the recent updates in the management of patients with sickle cell anaemia.*
Methods: *This review was done by conducting a search on Pubmed.gov and Google Scholar using the tag words "Sickle Cell Anaemia, Recent Management". Articles reviewed were those written within not more than ten years prior to the date of review.*
Conclusion: *Newborn screening for early detection, transcranial Doppler ultrasound, chronic transfusions, Hydroxyurea and haemopoietic stem cell transplantation are all new methods used in the management of this disease and lead to significantly reduced morbidity and mortality in patients with Sickle Cell Anaemia.*

INTRODUCTION
Sickle Cell Anaemia (SCA) belongs to the group of inheritable disorders known as Sickle cell disease, which are characterized by the presence of a sickle haemoglobin (HbS) caused by a mutation in the β-globin chain gene (βs).[1,2] SCA is the homozygous form of the disease (HbSS) and clinically the most severe Sickle Cell Disease (SCD) disorder associated with high morbidity and mortality especially within the first five years of life. However the disease exhibits great variety in the clinical presentation. The past two decades have seen advances in the treatment of SCA due to a better understanding of the pathogenesis of the disease.

SICKLE CELL ANAEMIA AS A GENETIC DISORDER
SCA was first diagnosed in an African American dental student in 1910 following presentation with anaemia, fever, jaundice and leg ulcers.[2] In Africa in the early twentieth century, there were reports of children in families dying at a very young age which may have been attributed to the disease.[3,4] In 1977, SCA became the first disease which the cause was attributed to a genetic mutation.[2] HbS arises due to a single nucleotide mutation where Thymine replaces Adenine in the sixth codon of the β-globin gene (GAG-> GTG). This results in a change in the amino acid produced; Valine, a non-polar amino acid replaces Glutamic Acid.[1,2,5] The variant haemoglobin produced (HbS) polymerizes easily into long crystals which elongate in conditions of reduced oxygen tension and cellular dehydration, changing the shape of the red cell from discoid to a sickled shape that is rigid and prone to both intravascular and extravascular haemolysis. This eventually leads to the hallmarks of this disease which include chronic haemolytic anaemia, vaso-occlusion and chronic organ damage.[2,6]

PATHOPHYSIOLOGY & CLINICAL MANIFESTATION
Knowledge of the pathogenesis of SCA helps in understanding the clinical presentation. It is now known that hypoxia occlusion in the microvasculature due to hypoxia is not the only pathophysiologic mechanism in this disease. SCA is a chronic inflammatory disease with acute episodes. Dysfunctional vascular endothelium, increased white blood cells, platelets, coagulation pathway, adhesion molecules and Nitrous Oxide metabolism derangements are all thought to play a role in

vaso-occlusion.[2] There is a variable phenotypic expression from person to person and even in the same individual over time. The exact reason for this is not known, but foetal haemoglobin (HbF), presence or absence of other genes (e.g. B-Thalassaemia) and environmental factors are thought to play a role.[7]

HbF has been found to be protective over HbSS disease and is an independent marker of outcome. Patients with a higher concentration of HbF have a milder form of the disease. HbF reduces in concentration afterbirth at about six months of age. From then on, the protection it offers ceases, making it possible for the child to manifest clinically with the disease.

The clinical presentation of SCA can involve virtually any organ in the body and is beyond the scope of this review. For the purpose of this article it can be broadly grouped into anaemia & its complications; vaso-occlusion & pain; and features that arise due to chronic organ damage.

There is a background chronic haemolytic anaemia with red cell lifespan reduced to about 17days (as against normal lifespan of 100-120 days) which patients with SCA are able to tolerate because HbS easily releases oxygen to the tissues and shifts the haemogobin-oxygen dissociation curve to the right. Due to ongoing haemolysis, the bone marrow is under constant stimulus for Erythropoiesis resulting in expansion of the marrow space involved in haemopoiesis and the typical sickle "facies". There are often sudden acute exacerbations of this chronic anaemia due to hyperhaemolysis, sequestration in organs, and aplastic crises due from parvovirus infection. Sequestration usually occurs in the spleen within the first five years of life after which most patients would have undergone fibrosis and functional asplenia (autosplenectomy).[2,7]

Vaso-occlusion is the hallmark of this disease in both children and adults, occurring in capillary and post-capillary venules following hypoxia and subsequent sickling of the red cells. Sickling can be precipitated by both intracellular and extracellular dehydration. Background endothelial dysfunction favours adherence of sickle cells to the vascular endothelium.

The potassium-chloride and gardos channels of the red cells have been implicated in cellular dehydration.[2,7] Nitrous oxide is normally synthesized by the endothelium and plays a role in vasodilation, while having anti-inflammatory and anti-platelet properties. In SCA, nitrous oxide is scavenged by free haemoglobin release by cell lysis. Nitrous oxide levels are reduced especially during Vaso-occlusive Crisis (VOC).[2]

Vaso-occlusion typically presents as bone pains involving the long bones. In the first year of life, it commonly manifests as hand-foot syndrome. Vaso-occlusion could also result in Acute Chest Syndrome (ACS), which is the commonest cause of death in adults. ACS presents as chest symptoms with fever and diagnosed with a chest x-ray showing a new pulmonary infiltrate. ACS usually occurs during VOC due to micro-embolization of fat from the bone marrow, which has undergone necrosis.

As people with SCA are living longer, the long term effect of SCA is coming into light. Any organ in the body can be affected. Table 1 gives a summary of organ involvement in SCA. Of all these, leading causes of significant morbidity and mortality are caused by stroke, renal insufficiency, chronic leg ulcers. Screening and prompt treatment of any organ involvement should be done routinely.

The lack of expression of the Duffy antigen in most African-Americans is associated with worse chronic organ involvement because this antigen on the red cell membrane promotes clearance of inflammatory cytokines.[8]

ROUTINE MANAGEMENT

Since the early 1960s the treatment of SCA has been mostly preventive, supportive and symptomatic-consisting of Folate therapy, prophylactic Penicillin from birth to about five years of age; vaccination encapsulated organisms, pain relief, hydration and blood transfusion.

Pain relief is dependent on the degree of pain the patient has and drugs used range from simple analgesics like Paracetamol, Non-Steroidal Anti-inflammatory agents (NSAIDs), Tramadol, opioids and opiates. The non-opioids have a ceiling effect, dose above which no additional analgesic effect is attained.[5] Paracetamol and NSAIDs can be given in addition to an opioid in very severe pain.[9]

Hydration is very important in preventing or reversing both intra- and extracellular dehydration. Blood transfusion is one of the mainstays of therapy. Transfusion may be episodic or chronic

(hypertransfusion). Indications for its use in SCA include sudden/ acute exacerbation of anaemia, Acute Chest Syndrome, prevention and treatment of stroke. Transfusion can be for the relief of an acute manifestation or can be chronic transfusions. It can also be simple or exchange blood transfusion (EBT). The type of transfusion given depends on the aim and indication for it. Generally the prevention and treatment of Stroke is by chronic transfusions with the aim of achieving a HbS concentration of <30% and suppressing endogenous production of sickle haemoglobin by the bone marrow by maintaining the haemoglobin concentration at about 10-11g/dl. Chronic blood transfusion is better given as exchange blood transfusion (EBT).[10] Short term EBT can be done to speed up healing of leg ulcers.[7] Acute chest syndrome is also treated with simple or exchange transfusion in addition to oxygen, antibiotics, pain relief, hydration and incentive spirometry.[5]

Transfusion of patients with SCA should be with HbAA blood, especially so when a low target HbS level is desired. Blood that has not been stored for long periods of time is preferred as these fresher red cells have a better capacity in oxygen delivery on getting to the tissues.

Transfusion is not without its complications, especially in the chronic setting. Apart from the infectious and non-infectious complications, multiply transfused sickle cell patients are mostly at risk for Iron overload, alloimmunization and also the development of autoantibodies. Iron overload is discussed below.

Alloimmunization develops when the patient produces antibodies to antigens that he or she lacks. This causes difficulty in crossmatching compatible units of blood for transfusion and predisposes to delayed haemolytic transfusion reaction.[10] Alloimmunization occurs in about 19-37% of patients[5,10] and increases with age and the number of units previously transfused.[9,10] The prevalence is significantly lower in Africa where it is estimated to occur in about 6.1% of SCA patients.[11] This prevalence is higher in western countries where majority of the donors are Caucasians and non-Africans donating to African-American patients, therefore with greater ethnic diversity there is greater antigen variation and higher risk of development of alloimmunization.[10,12,13] The transfusion of only compatible blood does not totally prevent alloimmunization.[13]

A systematic approach for the transfer of paediatric and adolescent sickle cell patients to adult clinics when they become adults is necessary.[14] Pregnant women with SCA should be treated as high risk pregnancy. Centres should have specific antenatal guidelines drawn up and adhered to strictly, maintaining close supervision in conjunction with the Haematologist. If the patient was on Hydroxyurea, it should be stopped immediately conception occurs. Delivery methods should ensure that patient does not get dehydrated.[15,16]

ADVANCES IN MANAGEMENT

Over the past ten to fifteen years, there has been progress made in the management of SCA. These advances are aimed at the prevention, disease modification and cure of SCA.

Newborn Screening

In Africa where the largest burden of SCA is, most patients are diagnosed at the first complication which makes them present to the hospital.[3] Newborn Screening is routinely done in the western world for the early detection and follow up management. This can be done using High Performance Lipid Chromatography (HPLC) or genetic analysis like Polymerase Chain reaction methods. Newborn screening is important because it allows for the early institution of preventive measures such as folate therapy, antibiotics and Hydroxyurea therapy which in the long run improves the prognosis.

Transcranial Doppler Ultrasound & Stroke Prevention

Stroke is a major cause of death especially in children less than ten years of age. About 11% of patients less than twenty years have had a stroke.[5] Patients who have had an incidence of stroke are more at risk to develop a recurrent stroke. Detection of the risk of stroke can now be determined by the use of systematic Transcranial Doppler (TCD) Ultrsound methods which assess the accelerated blood flow velocity of the cerebral arteries especially in children aged 2-16years. The use of TCD in predicting and preventing stroke in children has dramatically reduced the incidence of childhood stroke to about 2-3%.[5] Values >200cm/sec are abnormal, associated with a risk of 9% per year having a stroke and require treatment. Values between 170-199cm/sec have a conditional risk with 2-5% per year having a stroke. These patients require very close monitoring and preventive chronic transfusions. About 4.7% of children with HbSS have an abnormal TCD reading in Nigeria.[17] A patient at risk for stroke should be started on

chronic transfusions immediately as this has proven to reduce the occurrence and prevent the recurrence of stroke. Although chronic transfusions do not completely prevent a stoke from occurring or recurring, studies done show that those who do not receive chonic transfusion have a higher recurrence rate and mortality compared to those on chronic transfusion. In the management of stroke, HbS concentration should be targeted at <30% using HPLC and this is be better achieved by EBT rather than simple transfusions.

Iron Chelators

Daily iron requirement is between 1-3mg per day. A unit of blood contains about 200-250mg of iron. An individual transfused with two units of blood has more than a year's supply of iron, which is not easily excreted from the body. Multiply transfused sickle cell patients eventually develop iron overload. This excess iron is deposited in the liver, heart and other organs and contributes to long-term organ damage. Iron chelators used in the treatment of iron overload include Deferoxamine, Deferipone and Deferasirox. The first iron chelator, Deferoxamine, is given parenterally and is associated with several unwanted side effects, which include allergy at injection site, hearing loss, visual changes and growth impairment. It is not well tolerated by patients and this led to the development of the newer oral iron chelating agents Deferipone and Deferasirox. These are easier to administer and are better tolerated than Deferoxamine. Deferipone has a short half life and requires three doses a day, while Deferasirox has a longer half life is taken as a daily dose.[10]

HbF inducers- Hydroxyurea

The clinical variability of the phenotypic expression in patients with SCA is largely dependent on the concentration of HbF. This led to the search for inducers of HbF. Hydroxyurea (also known as Hydroxycarbamide) is a chemotherapeutic agent that inhibits Ribonucleotide Reductase, an enzyme which is required for DNA synthesis.[1,18] It is a potent inducer of HbF and has been established as a disease-modifying drug in the treatment of HbSS disease. The mechanisms by which Hydroxyurea (HU) acts in SCA are multiple and still under study. HU induces stress Erythropoiesis, which increases the pool of HbF producing erythrocytes. Other possible mechanisms of action in SCA include increased nitrous oxide production, macrocytosis with increased cellular hydration and altered expression of adhesion molecules.[1] The mechanisms of action of HU are shown in Fig.1.

Hydroxyurea reduces the frequency and severity of VOC, improves overall blood flow and has also been shown to reduce transfusion requirements, prevent recurrence of ACS and mortality of sickle cell patients on HU is significantly reduced.[1,7,10,18] Since the onset of its use in the treatment of SCA in 1984, HU has been shown to reduce the overall mortality and morbidity of the disease. When the drug was first approved for use in SCA, patients had to satisfy inclusion criteria which included three severe VOC crises or two ACS per year leading to three or more hospital admissions per year.[19] Recently, HU is being recommended as the standard of care for SCA patients regardless of age or clinical severity.[1] HU was also initially given to only adults, but now HU is given to children with SCA from even as early as nine months of age in western countries.[20] Children and adults have similar efficacy with the drug.[18]

The dose range of HU is 15-30mg/kg. A starting dose of 15mg/kg is given, which is then escalated by 5mg/kg every 8-12 weeks until the maximum tolerated dose of 35mg/kg is reached or toxicities develop.[1,19] Dose escalations should be done every 12 weeks while monitoring full blood count and other parameters initially weekly, then bi-monthly and eventually monthly. HU therapy causes dose-dependent increase of the red cell mean corpuscular volume (MCV) and Haemoglobin concentration while reducing the total reticulocyte count, total white cell count (WBC) and Lactate Dehydrogenase (LDH).[1,7,18] The baseline MCV should be done before commencing HU and this can be used to monitor compliance to therapy.

Side effects of HU are Neutropenia, thrombocytopenia, anaemia and deranged liver functions test. These toxicities are dose dependent and HU can be suspended and recommenced at a lower dose when toxicity resolves.[19] Skin discolouration, hyperpigmentation and painful leg ulcers also occur. These are known side effects seen when HU is used in treatment of myeloproliferative disorders. The presence of leg ulcers is not an absolute contraindication to its use, however, painful leg ulcers have not been reported in sickle cell patients receiving HU.[1,7] There have been concerns about the long term safety of the drug, since it interferes with DNA synthesis and repair.[18] SCA patients who have been on long term therapy are not more at risk in developing leukaemias or myelodysplastic syndromes than the general public,[10,18] however the long term safety profile remains a concern.[1]

Hydroxyurea therapy is absolutely contraindicated in pregnancy, and young adults on the drug should be on contraception. In the event of pregnancy, the drug must be immediately discontinued die to potential teratogenicty.[15,16]

Haematopoietic Stem Cell Transplantation (HSCT)

The haematopoietic stem cell is capable of re-constituting haemopoiesis in a lethally irradiated host. HSCT involves the total or subtotal ablation of the bone marrow using chemotherapy at maximally tolerated doses (with or without irradiation), and then re-constituting haemopoiesis with stem cells, with the aim of achieving cure for a malignant or non-malignant condition likeSickle cell anaemia. Matched sibling donors are best used, but this may not be possible since most sickle cell patients will have siblings with the sickle trait.The first report of HSCT for SCA was done in a sickle cell patient with Acute Myeloid Leukaemia, whose genotype changed from HbSS to HbAS and this brought to light the fact that HSCT can be used to achieve a cure.[18,21,22]

Haematopoietic Stem Cell Transplantation is offered to younger patients who have severe disease. Presently, HSCT is the only potential cure for SCA presently. Successful transplant changes the host's haemoglobin genotype from HbSS to HbAS or HbAA, depending on the donor's genotype and if mixed chimerism is present. It should however be emphasized that HSCT does not modify the genes of the host and therefore these patients will still transfer the sickle gene to their offspring. They should undergo genetic counseling. HSCT has recently commenced successfully in Nigeria for the treatment of SCA and patients who will benefit from HSCT and can afford it can be referred for the procedure.

OTHER THERAPIES

As the understanding of the Pathophysiology of SCA improves, potential therapeutic targets are developed. Table 2 gives an overview of potential therapeutic targets, some of which are still being researched.

Priapism can be managed with the intracavernous injection of the a-adrenergic agonist, Etilefrine. There is an ongoing clinical trial of Arginine Butyrate to accelerate healing of leg ulcers which appears promising.[7]

Niprisan is a drug produced from four plant extracts by the National Institute for Pharmaceutical Research and Development (NIPRD) in Nigeria that demonstrated clinical efficacy and safety. The drug has potent anti-sickling properties and was shown to reduce VOC. It was initially launched in Nigeria in 2006 following NAFDAC approval. However, financing for the production of the drug even after collaboration with a United States drug company was poor. Eventually in 2009, the drug's license was withdrawn after the US drug company filed for bankruptcy.[23] The NIPRD has also identified other plants with anti-sickling properties in Nigeria and research is still ongoing.[4]

THE BURDEN OF SICKLE CELL ANAEMIA

About 2% of Nigerians are born with SCA.[17,24] The World Health Organization (WHO) estimates that about 4 million people in Nigeria have SCA with 150,000 children born annually with the disease. SCA accounts for up to 16% of under five year mortality in Nigeria, but life expectancy has improved dramatically for SCA patients in western countries.[25]Nigeria has a 24% carrier rate of the sickle cell trait. It is ironical that the UK and US, which bear only about 1% of the disease burden, are where most of the research and recent advances in therapy of SCA have been done. Therapeutic advances in the treatment of SCA have not yet been implemented on a wide scale in our country and therefore no significant impact on the morbidity and mortality associated with this disease has been achieved.[3,25] The prevalence of SCA may actually be underestimated because numerous people in the rural setting have no access to proper health care facilities. Traditional and religious beliefs also get in the way of patients presenting to the hospital, as sickle cell is perceived in some rural areas as an incurable disease.[4] Many patients are diagnosed when they first present with a complication at the hospital because they were not screened at birth. The management of SCA is a lifelong process and therefore constitutes an economic burden on affected families.

PREVENTION

Education of the general public and Genetic Counseling of at risk couples cannot be over emphasized. Screening methods for carriers of the sickle cell trait are inexpensive and readily available. Identification of couples at risk and prenatal diagnosis by chorionic villus sampling from nine weeks of gestation can be done.
National control programmes and a systematic screening approach need to be put in place.[3,25]

SUMMARY

The management of SCA is multi-faceted, multi-disciplinary and life-long from the time the diagnosis is made. Preventive and palliative measures involve the use of folic acid, penicillins, vaccinations, pain relief, hydration and transfusion. Newborn screening is recommended for the early detection and follow-up of these patients. Chronic transfusion is required to treat and prevent stroke. Hydroxyurea is a disease-modifying drug that mainly acts by increasing the concentration of HbF and this in turn ameliorates the clinical features of the disease and reduces the overall morbidity and mortality. HSCT has successfully cured patients with HSCT. Gene therapy is still under investigation for the cure of SCA.

CONCLUSION

The burden of SCA in Nigeria can be greatly reduced by the implementation on a wide scale of these new methods in management of SCA. Measures to increase awareness of SCA by both the general public and the government should be put in place, as this will go a long way for the control of this disease in a country with such a large burden of SCA. Newborn screening should commence in at least every major referral centre in the country. Hydroxyurea is readily available and should be used in treating both adults and children with SCA as this improves outcome of the disease.

The government should be actively involved to provide better health care facilities so that procedures like Transcranial Doppler ultrasound and other new methods of management can be made readily available for patients with SCA.

REFERENCES

1. McGann PT, Ware RE; Hydroxyurea for Sickle Cell Anaemia: What have we learned and what questions remain? Current Opinion in Hematology 2012, 18:158-165
2. Kaushansky K, Lichtman M, Beutler E, Kipps J.T, Seligsohn U, Prchal J; Disorders of Hemoglobin Structure: Sickle Cell Anemia & Related Abnormalities in Williams Hematology, Chapter 48, McGraw-Hill 8th ed, 2010, p 871-914, 2010
3. Diallo D, Tchernia G; Sickle cell Disease in Africa. Current Opinion in Hematology 2002, 9:111-116
4. Ameh SJ, Tarfa FD, Ebeshi BY; Traditional Herbal Management of Sickle Cell Anaemia: Lessons from Nigeria. Anaemia Vol. 2012, Article ID 607436, 9 pages
5. Ballas SK, Kesen MR, Goldberg MF, Lutty GA, Dampier C, Osunkwo I. et al; Beyond the definitions of the phenotypic complications of sickle cell disease: An update on management. The Scientific World Journal Vol.2012, Article ID 949535, 55 pages
6. Malowany JI, Butany J; Pathology of Sickle Cell Disease. Seminars in Diagnostic Pathology 2012, 29; 49-55
7. Bartolucci P, Galcteros F; Clinical management of adult sickle cell disease. Current Opinion Hematology 2012, 19:149-155
8. Afenyi-Annan A, Kail M, Combs MR, Orringer EP, Ashley-Koch A, Telen MJ; lack of Duffy antigen expression is associated with organ damage in patients with sickle cell disease. Transfusion 2008; 48:917-924
9. Gillis VL, Senthinathan A, Dzingina M, Chamberlain K, Banks E. et al; management of an acute painful sickle cell episode in hospital: Summary of the NICE guidance. British Medical Journal 2012; 344:e4063
10. Smith-Whitley K, Thompson AA; Indications and Complications of Transfusions in Sickle Cell Disease. Pediatrics Blood Cancer 2012; 59:358-364
11. Natukunda B, Schonewille H, Ndugwa C, Brand A; Red cell alloimmunization in sickle cell disease patients in Uganda. Transfusion 2010; Vol.50, 20-25
12. Chou ST, Liem RI, Thompson AA; Challenges of alloimmunization in patients with haemoglobinopathies. British Journal of Haematology, 2012, 159, 394-404
13. Yazdanbakhsh K, Ware RE, Noizat-Pirenne F; Red cell alloimmunization in sickle cell disease: Pathophysiology, risk factors and transfusion management. Blood 2012, 120:528-537
14. DeBaun MR, Telfair J; Transition and sickle cekll disease. Paediatrics 2012; 130; 926
15. Howard J, Oteng-Ntim E; The Obstetric management of Sickle cell disease: Best Practice and Research Clinical Obstetric and Gynaecology 26(2012) 25-36
16. Oteng-Etim E, Nazir S, Singhal T, Howard J; Sickle cell disease in pregnancy. Obstetrics Gynaecology and Reproductive Medicine 2012 22:9, 254-263
17. Ware RE; How I use Hydroxyurea to treat young patients with sickle cell anaemia. Blood 2010; 115:5300-5311
18. Hydroxyurea Protocol: Sickle Cell Information Center
19. Wang WC, Ware RE, Miller ST, Iyer RV, Casella JF, et

al; A multicenter randomized controlled trial of Hydroxyurea (hydroxycarbamide) in very young children with sickle cell anaemia. Lancet 2011 May 14; 377 (9778): 1663-1672
20. Vermylen Ch, Cornu G, Philippe M, Ninane J, Borja A, Latinne D. et al; Bone marrow transplantation in sickle cell anaemia. Archives of Disease in Childhood 1991; 66:1195-1198
21. Billings FT; Treatment of Sickle Cell Anaemia with Bone marrow transplantation- Pros and Cons. Trans Am Clin. Climatol. Assoc. 1990:101;8-20
22. Perampaladas K, Masum H, Kapoor A, Shah R, Daar AS, Singer PA; The road to commercialization in Africa: Lessons from developing the sickle cell drug Niprisan. BMC International Health and Human Rights 2010, 10(Suppl1): S11
23. Ogunrinde GO, Keshinro MI, Ige SO; HIV seropositivity in children with sickle cell disease. Annals of African Medicine. 2005. Vol.4, No 3: 104-106
24. Lagunju I, Sodeinde O, Telfer P; Prevalence of Transcranial Doppler abnormalities in Nigerian children with Sickle cell disease
25. World Health Organization: Sickle Cell Anaemia Report by Secretariat. 2006, Geneva