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In

Clinical Haematology

For the examinations to be conducted in February 2016

Department of Clinical Haematology

Christian Medical College, Vellore.

Tamil Nadu, India.

Clinical and Demographic Profile in Patients with Sickle Cell Disease: A single Centre Study from India.

CERTIFICATE

This is to certify that this thesis titled "Clinical and Demographic Profile of patients with Sickle cell disease: A single Centre study from India" is a bonafide work of the candidate, Dr. Reashma Roshan during the period from August 2012 to July 2015 in partial fulfillment, towards the award of degree of Doctorate of Medicine (higher specialty) in Clinical Haematology for the examinations to be conducted by the Dr.M.G.R Medical University in February 2016.

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(In the name of God, Most Gracious; Most Merciful)

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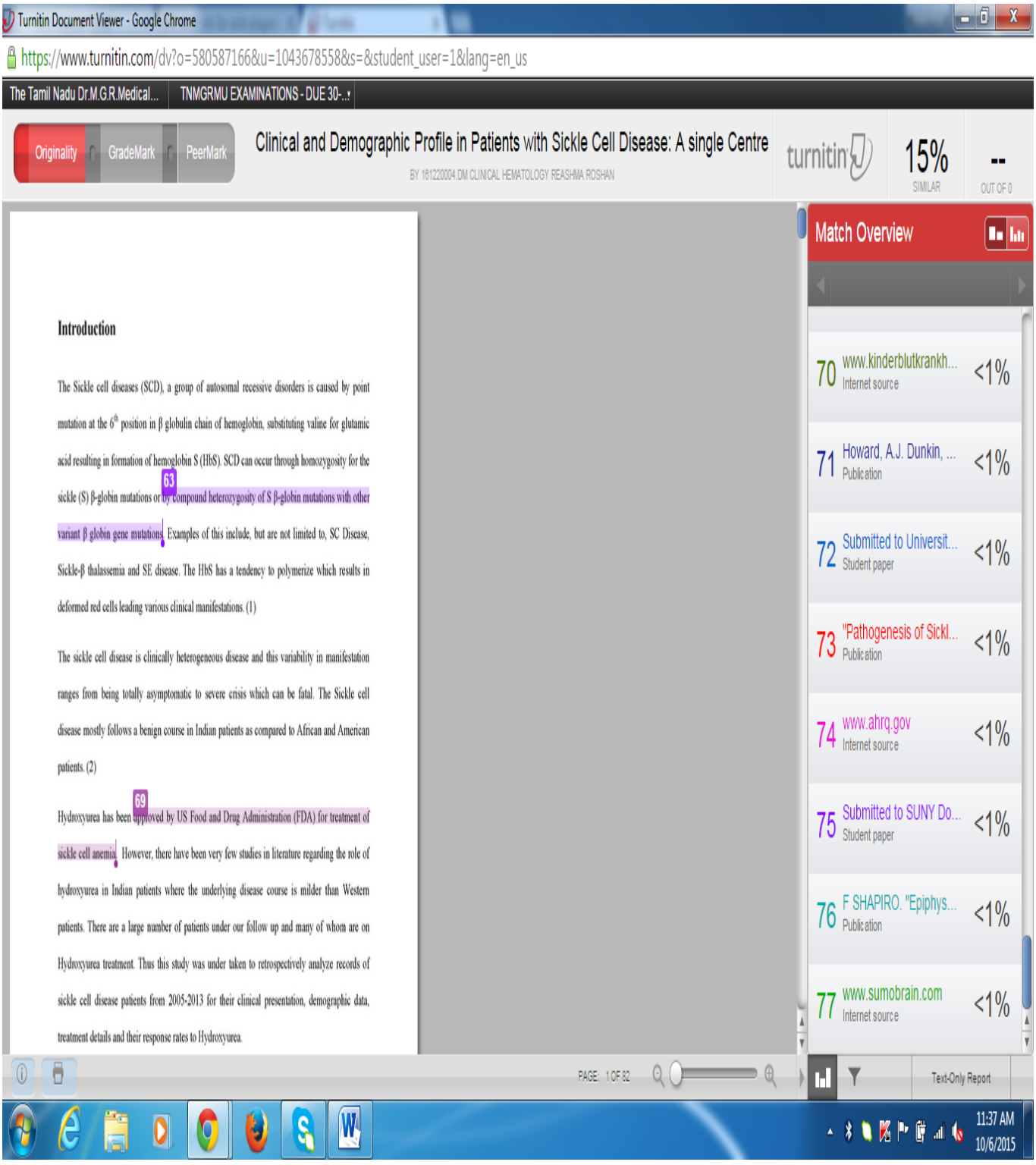
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Introduction
The Sickle cell diseases (SCD), a group of autosomal recessive disorders is caused by point mutation at the 6th position in β globulin chain of hemoglobin, substituting valine for glutamic acid resulting in formation of hemoglobin S (HbS). SCD can occur through homozygosity for the sickle (S) β-globin mutations or by compound heterozygosity of S β-globin mutations with other variant β globin gene mutations. Examples of this include, but are not limited to, SC Disease, Sickle-β thalassemia and SE disease. The HbS has a tendency to polymerize which results in deformed red cells leading various clinical manifestations. (1)
The sickle cell disease is clinically heterogeneous disease and this variability in manifestation ranges from being totally asymptomatic to severe crisis which can be fatal. The Sickle cell disease mostly follows a benign course in Indian patients as compared to African and American patients. (2)
Hydroxyurea has been approved by US Food and Drug Administration (FDA) for treatment of sickle cell anemia. However, there have been very few studies in literature regarding the role of hydroxyurea in Indian patients where the underlying disease course is milder than Western patients. There are a large number of patients under our follow up and many of whom are on Hydroxyurea treatment. Thus this study was under taken to retrospectively analyze records of sickle cell disease patients from 2005-2013 for their clinical presentation, demographic data, treatment details and their response rates to Hydroxyurea.
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Sub: **Fluid Research Grant Project:**
Clinical and demographic profile of sickle cell disease patients: A single centre experience from India.
Dr. Reashma Roshan, PG Registrar, Dr. Biju George, Dr. Alok Srivastava, Dr. Vikram Mathews, Dr. Auro Viswabandya, Dr. Aby Abraham, Dr.Abhijeet. P Ganapule, Dr. Fouzia N A, Clinical Haematology, CMC, Vellore.

Ref: IRB Min No: 9001 [OBSERVE] dated 04.08.2014

Dear Dr. Reashma Roshan,

The Institutional Review Board (Blue, Research and Ethics Committee) of the Christian Medical College, Vellore, reviewed and discussed your project entitled "Clinical and demographic profile of sickle cell disease patients: A single centre experience from India." on August 4th 2014.

The Committees reviewed the following documents:

1. IRB Application format
2. Curriculum Vitae' of Drs. Reashma Roshan, Biju George, Alok Srivastava, Vikram Mathews, Auro Viswabandya, Aby Abraham, Abhijeet. P Ganapule, Fouzia N A.
3. No of documents 1-2

The following Institutional Review Board (Blue, Research & Ethics Committee) members were present at the meeting held on August 4th 2014 in the CREST/SACN Conference Room, Christian Medical College, Bagayam, Vellore 632002.

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We approve the project to be conducted as presented.

The Institutional Ethics Committee expects to be informed about the progress of the project, any **adverse events** occurring in the course of the project, any **amendments in the protocol and the patient information / informed consent**. On completion of the study you are expected to submit a copy of the **final report**. Respective forms can be downloaded from the following link: http://172.16.11.136/Research/IRB_Polices.html in the CMC Intranet and in the CMC website link address: http://www.cmch-vellore.edu/static/research/Index.html.

Yours sincerely

Dr. Nihal Thomas
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Cc: Dr. Biju George, Haematology, CMC, Vellore.

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Introduction

Introduction

The Sickle cell diseases (SCD), a group of autosomal recessive disorders is caused by point mutation at the 6^{th} position in β globulin chain of hemoglobin, substituting valine for glutamic acid resulting in formation of hemoglobin S (HbS). SCD can occur through homozygosity for the sickle (S) β-globin mutations or by compound heterozygosity of S β-globin mutations with other variant β globin gene mutations. Examples of this include, but are not limited to, SC Disease, Sickle-β thalassemia and SE disease. The HbS has a tendency to polymerize which results in deformed red cells leading various clinical manifestations. (1)

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Review of literature

Review of Literature

The relevant literature regarding sickle cell disease will be reviewed as follows

I. Definition and classification

II. Epidemiology

III. Genetic Basis and Pathophysiology

IV. Clinical Profile and Complications

V. Phenotypic Heterogeneity and Genetic modifiers

VI. Management of Sickle cell disease and Role of Hydroxyurea

I. Definition and Classification:

In 1910, James Herrick, for the first time recognized a clinical entity with symptom complex of anemia, recurrent fever and bouts of bone pains associated with sickle shaped red blood cells which was subsequently named as Sickle cell anemia.(2)

The term sickle cell disease is used for all different genotypes that cause characteristic clinical syndrome whereas sickle cell anemia is usually used to refer specifically to homozygous HbS disease (HbSS). The various genotypes that can be associated with SCD phenotype include HbS/Beta thalasemia, HbSC and other rarer genotypes like HbS/HbE , HbS/HbD etc.(3)

II. Genetic basis and Pathophysiology

Hemoglobin S (HbS) is formed as result of mutation in beta globulin gene in which 17^{th} nucleotide is changed from thymine to adenine which leads to substitution of valine instead of

glutamic acid at sixth amino acid in beta globulin chain. This mutation produces a hydrophobic motif in deoxygenated HbS. The SCD phenotype can occur in both homozygous state or in compound heterozygous states in which HbS is combined with other mutation on second beta globulin gene such as beta thalasemia, HbC, HbD , HbE etc.(3)

In 1949, Linus Pauling and Colleauges discovered sickle hemoglobin (HbS) due to its differential electrophoretic mobility on gel electrophoresis and this was first demonstration of an abnormal protein linked to genetic disorder. Thus the term "molecular disease" was coined and sickle cell disease was the first example. (4)

In ensuing decades, detailed pathophysiology of SCD was described based on abnormal polymerization of deoxygenated HbS. More recent data has indicated the role of endothelial activation with increased adhesion, vasoconstriction and coagulation activation in pathogenesis of SCD.

POLYMERIZATION OF HbS

The concentration of Hb within RBCs is very high (32-34g/dl) and this requires hemoglobin molecule to be highly soluble. After deoxygenation of HbS, the replacement of hydrophilic glutamic acid with hydrophobic valine results in interaction with another deoxygenated HbS molecule, triggering an aggregation which leads formation of large polymers. The repeated cycles of oxygenation and deoxygenation of RBCs produce repeated sickling and unsickling, leading to RBC damage and increased rate of red cell breakdown as well as short-lived sub-populations of very high density irreversibly sickled RBCs. This polymerization of deoxygenated HbS is the primary event in the pathogenesis of sickle cell disease, which results in a distortion of the shape of the red cell with marked decrease in its deformability. These rigid red cells are

responsible for the vaso-occlusive phenomenon which is considered the hallmark of the disease. (5)

The polymerization of HbS is a dynamic event and kinetic features of polymer formation are determinants of the shape and morphology of red cells. If deoxygenation is rapid, multiple independent polymerization events occur, that result in a granular or cobblestone texture which does not alter the cell's disk-like shape. In contrast, when sickle red cells are slowly or partially deoxygenated, there is formation of single nucleus of aggregated molecules of deoxygenated hemoglobin S. This nucleation is leads to growth and alignment of fibers, which transforms the cell into a classic sickle shape. This distortion of the shape of the red cell by projections of aligned hemoglobin S fibers has an important role in disturbing the structure and function of the membrane in sickle red cells, enhanced in part by oxidant stress (6,7).

The rate and amount of polymer formation in a circulating sickle red cell depend primarily on three factors: the degree of deoxygenation, the hemoglobin concentration in RBC, and the presence or absence of hemoglobin F. The lag period required for the formation of polymer is designated as the delay time which is inversely proportional to the HbS concentration in the red cell. As the range of transit times in the microcirculation is usually short as compared to the range of delay times for sickle red cells, polymers are not formed in most of the red cells (over 80 percent) during their flow through the arterioles and capillaries.(8)

The formation of polymer fibers in sickled RBCs triggers cascade of other cellular abnormalities involved in overall pathophysiological mechanisms. Dysregulation of cation homeostasis results from the activation of various ion channels including the K-Cl co-transport system and the Ca-dependent K-channel (Gardos channel) in particular, which leads to a loss of potassium resulting

cellular dehydration. Because of cellular dehydration, the intracellular Hb concentration increases, favoring deoxy-HbS polymerization. The Hb in sickled red cells becomes denatured and hemichromes are concentrated at the internal side of the membrane along with proteins of the cytoskeleton, particularly protein band 3. This process also involves the loss of heme and the release of Fe^{3+} which promotes the formation of an oxidizing microenvironment. This leads to disruption of normal asymmetry of cell membrane with exposure of phosphatidylserine at cell surface. Due to exposure of Band 3 protein, Anti-band 3 IgGs accumulates on the protein band 3 aggregates, leading to erythrophagocytosis by macrophages. Finally, all these membrane changes lead to the production of micro particles. (9, 10)

As RBCs are the predominant cell type in blood, so their characteristics largely determine the rheologic and hemodynamic behavior of blood. The interactions of the RBC membrane skeleton and membrane proteins with the lipid bilayer provide the flexibility for RBC to deform under shear stress in circulation and to regain its shape as a biconcave disk. In SCD, formation of hemoglobin polymers in the sickled RBC affects the RBC's ability to maintain its normal morphology especially during shear stress. The recurrent episodes of sickling form stiff and fragile RBC, which in turn leads to vasoocclusion and hemolytic anemia respectively. (11)

However these mechanisms do not explain the events that triggers VOCs. As explained earlier, during basal conditions, the delay time which is necessary for the polymerization of deoxy HbS is usually longer than the time required for of passage of RBCs in microcirculation. Recent data provide additional evidence on various mechanisms including slowing down the blood flow in the microcirculation, are likely to trigger VOCs. (10)

Role of Vascular Endothelium

The polymerization of deoxy HbS which subsequently leads to stiffening and fragility of sickled red cells explains the pathogenesis of vasoocclusive crises and hemolytic anemia noticed in SCD. However, it does not to explain the triggering of vasoocclusive crisis as under basal condition, the time required for sickling (referred as delay time) is usually more than time of passage of RBCs in microcirculation. Accordingly, various studies were carried out to study the interaction between SS red cells and vascular endothelium and the factors that can lead to slowed passage of RBCs in microcirculation which can trigger polymerization within RBCs and ultimately precipitate VOC.

During 1980s, the team of R.P. Hebbel showed that there is an increased adhesion of the sickled RBCs to the endothelium that delays their passage through microcirculation.. The study revealed that the main cells responsible for this abnormal adhesion are a population of young RBCs, called as “stress reticulocytes”. These stress reticulocytes, come out prematurely from the bone marrow due to anemic stress and express on their surface certain adhesion proteins that are normally required for their homing in the marrow.(12)

The first molecules identified responsible for this abnormal interactions on RBCs were the α4β1 integrin , very late antigen-4 (VLA-4) which directly binds both to fibronectin and the vascular cell adhesion molecule-1 (VCAM-1) which is expressed on the endothelial surface, especially after activation. In addition, CD36 which is expressed both sickled red cells and endothelium, interact with each other through a molecular bridge formed by a molecule of thrombospondin (TSP) secreted by activated platelets. In addition, numerous other receptor/ligand couples have been subsequently identified on the red blood cells, on one side, and on the endothelial cells on

the other, and involvement of various plasma proteins in addition to TSP, with description of intricate network of probably co-operative as well as redundant interactions. The situation varies differs to the vascular territories, e.g, VCAM-1 is specific for the endothelial cells of the microcirculation whereas von Willebrand factor mediates abnormal cell-cell adhesions in large vessels. (13)

The ongoing endothelial activation is evident as number of circulating endothelial cells(ECs) showing increased expression of E-Selectin, P-Selectin, ICAM-1 and VCAM-1 are present in sickle cell patients. The role P-Selectin has been especially studied in detail in murine models and its pivotal role in pathogenesis of vasocclusive crisis has been explained in knockout mice models.(14)

Sickle cell Vasculopathy

For decades, vaso-occlusion by rigid sickled red cells leading to tissue infarction was considered the only cause of organ dysfunction in patients with SCD. In 1991, Ballas et al, suggested two distinct sub phenotypes of clinical complications in SCD , one with recurrent painful crises and the other with recurrent ulcerations. (15). Furthermore, studies by Duits and Schnog expanded concepts of vascular dysfunction and epidemiologic studies from Serjeant and colleagues in 2004 further refined Balla's concept of SCD sub phenotypes.(the leg ulcer versus painful crisis).(16)

In the last decade, Gladwin and his colleagues outlined the importance of haemolysis in pathophysiology of SCD. In his study, Gladwin and his colleagues demonstrated due to intravascular hemolysis, hemoglobin and arginase are released from the red cells into plasma, where they scavenge nitric oxide (NO) as well as its precursor L-arginine. This reduces NO

bioavailability, especially in those patients with the high rate of hemolysis. The depletion of No is associated with vasoconstriction as well as endothelial dysfunction. The chronic NO depletion has also been implicated in development of vascular proliferation, pulmonary hypertension and activation of endothealial cells and platelets as well as in release of potent vasoconstrictor Endothelin.(16)

Subsequently, Kato and Gladwin showed that subgroup of SCD patients have high intravascular hemolysis, with decreased nitric oxide bioavailability, pulmonary hypertension, priapism and leg ulceration. Combining their own data with Steinberg and publications by others, a vasculopathy subphenotype was formulated by them comprising pulmonary hypertension, priapism, leg ulceration and preliminarily stroke, versus a viscosity-vaso-occlusive subphenotype involving vasoocclusive pain crisis, the acute chest syndrome and osteonecrosis.(17)

The role of Polymorphonuclear neutrophils (PMN) in the precipitation of VOC and has also been described in last decade.H yperleukocytosis is very common finding in SCD patients, and a high PMN count is a has been shown to negatively impact overall survival in SCD. The presence of adherent leukocytes in small post-capillary veinules suggests that leukocytes, because of their cell volume, are major participants in slowing down the microcirculation that initiates VOCs. In addition, invitro studies have shown that SS-RBCs can interact with leukocytes and particularly with PMNs.(18)

Coagulation Activation

The SCD is considered as hypercoagulable state due to multiple factors. There is chronic depletion of NO, which leads to activation of platelets as well as release of many procoagulant factors. As phosphatidylserine is abnormally exposed at the surface of sickled RBC and tissue

factor is expressed by activated circulating ECs and monocytes, these together participate to a borderline activation of the coagulation system. This leads to generation of thrombin, albeit at minimal level, which in turn exacerabrates the underlying ischemia-reperfusion injury associated with vasoocclusive crises.

In summary pathogenesis of sickle cell disease is driven by the principles of hemoglobin S polymerization within red RBCs, modulated by factors like inflammation and cellular adhesive events in the microcirculation which leading to ischemia and reperfusion events. This is amplified by intravascular hemolysis with deprivation of NO, activation of coagulation, increasing the expression of critical adhesion molecules on RBCs and endothelium along with leukocytes, and platelets that leads to sickle cell vasculopathy resulting in chronic organ dysfunction in patients of SCD.

III. Epidemiology:

SCD is one of the most common inherited blood disorder affecting human beings. It is believed that sickle Hb originated in Africa and population migration is responsible for wide spread dissemination of the gene. In addition, India is considered the place of origin. It is estimated that 250,000 children are born annually with SCD worldwide and is among the most important epidemiological genetic diseases in the world.(19)

In Africa, where disease has originated, the carrier frequency is about 10-30% in some regions. It is estimated that 200,000 new cases of SCD occur each year in Africa and more 70% of SCD patients live in Africa and is identified as a problem of major public-health significance.(Q)

In US, SCD is the most common inherited blood disorder with about 70,000 people living with SCD. It is more common in African-American and occurs 1 in every 500. The yearly estimate of affected births in North America is 2600. (2)

In UK, SCD affects 1 in 1300 live births, and about 12,000 individuals are living with SCD, making SCD the most common and fastest-growing genetic disorder in the UK. (2,20)

In 1952, Lehman and Cutbush first time detected Sickle gene among the tribal population of

Nilgiri Hills and almost at same time Dunlop and Mazumder detected five cases of sickle

Cell trait and three presumptive cases of sickle cell anemia among labourers in Assam, originating from tribal populations of Bihar and Orissa . Subsequent studies have revealed a high prevalence of sickle cell disease in India. The highest prevalence has been recorded in the Orissa (1-44.4%), followed by Madhya Pradesh (1-40.0%; including Chhattisgarh), Tamil Nadu (1-40.0%), Andhra Pradesh (1-35.7%), Assam (1-35.5%), Maharashtra (0.8-35.0%), Gujarat (1-31.4%),Kerala (1-30.0%), Uttar Pradesh (1.5-18.5%), Karnataka (1-8.0%), Rajasthan (1-5.7%), West Bengal (1-1.7%), and Bihar (0.8%; including Jharkhand).(21)

Although the exact incidence and prevalence of SCD is not known in India, due to lack of population based registry in majority of places, SCD still represents a significant public health problem in India. With a population of over 1.2 billion individuals, it is estimated that India is home to over 50% of world's SCD patients and the annual number of newborns in India with SCA was estimated at 44,000 .(22)

IV. Clinical Profile and Complications

There is marked heterogeneity in clinical presentation in patients with sickle cell disease ranging from totally asymptomatic to life threatening condition like acute chest syndrome. More common presentations include musculoskeletal pain due to occlusive crisis and increased weakness due to symptomatic anemia. The rarer presentations include avascular necrosis of head of femur, priapism, non-healing ulcer and acute chest syndrome. The physical examination is may reveals moderate splenomegaly usually in cases of compound heterozygous HbS/Beta thalassemia cases, along with mild pallor. The veno occlusion and hemolysis, which are the hall mark of SCD lead to major clinical manifestation and various complications and are described below.

1.Acute painful episodes (Veno occlusive Crisis):

Acute pain is the first symptom of the disease in 25% of patients and recurrent episodes of acute pain, also known as veno-occlusive episodes or veno occlusive crisis (VOC) is the most common manifestation of the disease. VOC is defined as the occurrence of pain in the extremities, back, chest, abdomen, or head that lasts two or more hours and is considered hall mark of SCD.(23)

The pain in hands and feet (known as dactylitis) is the most common symptom in children who present before two years of age and occur in 50% of children with SCD. The frequency of painful episodes usually peaks between 19-39 years and in patients. In a study by Platt OS et al, it was shown that patients aged above 20 years, higher frequency of acute painful episodes was associated with higher mortality.(24)

There is considerable variability in frequency and severity of painful episodes. Some patients may have six episodes annually while others may have fewer than three episodes and still many

patients do not have painful episodes at all. In a study by Cooperative Study of Sickle Cell Disease , the mean number of pain episodes/ year was 0.8 in SCA, 1.0 in sickle cell-beta (0) thalassemia, and 0.4 sickle cell-beta (+) thalassemia. Among the patients with SCA, 40 percent had no painful episodes, while 1 percent had 3 to 10 episodes/ year.(24,25).

The acute painful episodes usually affect long bones and joints, and the low back is the most frequently reported site of pain .In children, pain in hand and feet is usually the presenting manifestation. Other parts of the body which may be involved include the scalp, face, jaw, abdomen, and pelvis. The occurrence of three or more of acute painful episodes in year indicates that the patient has severe SCD. The severe abdominal pain can present as acute abdomen. (25).

Most studies have studied the pain frequency based on hospital emergency department visits. However, Smith WR et al studied the pain frequency as reported by patients on daily basis to assess the frequency of self-reported pain. They concluded that pain was reported in 55 percent of analyzed patient days and 29 percent of patients had pain almost daily. In this study, a painful episode without using hospital care was the" rule rather than the exception" and concluded that previously reported frequency of painful crises is an underestimate.(26)

The various factors that may precipitate pain episodes include dehydration, weather conditions, stress, infection, menses and rarely obstructive sleep apnea. However, in most cases, no precipitating factor may be identified. In about half of the pain episodes, clinical signs such as fever, swelling, tenderness, tachypnea, nausea, and vomiting is present.(27)

There are no diagnostic test to diagnose a vaso-occlusive episode and diagnosis is usually made on the basis of clinical features .The peripheral blood smear may reveals 5 to 50 percent of the red cells are irreversibly sickled RBCs . The most useful laboratory indicators of acute episode of

VOC are changes in density distribution of sickled RBC subpopulations and the rheologic properties of the blood, but tests may not be readily available. The pain episode is also associated with changes in levels of acute phase reactants (eg, C-reactive protein, fibrinogen) , serum lactate dehydrogenase, interleukin-1, tumor necrosis factor, and serum viscosity.(28)

In addition to acute painful episodes, many patients in SCD experience chronic low level pain, mainly in bones and joints, with intermittent exacerbation as acute episodes.

2. Anemia

Sickle cells are mechanically weak and are prone to hemolysis leading to anemia. However, as it is chronic, it is usually well tolerated. In a study by Embury SH et al, the mean hemoglobin in patients with SCD was 7.9g/dl. The anemia is usually more severe in homozygous state while variants of SCD patients are usually less anemic. The other factors which may worsen the the underlying chronic anemia is folate deficiency if not appropriately replaced.(29)

There are some complications of SCD which can lead to rapid drop in hemoglobin with resultant acute severe anemia. These include splenic sequestration, aplastic crisis and hyper hemolytic crisis and will be discussed separately.

3. Neurological complications

The neurological complications are one of the major complications in SCD. In patients of SCD, about 24% experience and overt clinical event like stroke by 45 years of age and in children, about 25% have silent ischemic changes that may impair neurocognitive function. In both children and adults, cerebrovascular events are one of the leading causes of mortality.(30,31)

The CNS complications in SCD include cerebral infarct, intracranial hemorrhage as well as cognitive and behavioral abnormalities. In a study by Cooperative Study of Sickle Cell Disease

(CSSCD), the incidence of CVA in homozygous sickle cell anemia was 0.61 per 100 patent years while it was0.08-0.09 in sickle beta thalassemia patients.(30)

The cerebral infarcts are more common in children with SCD as compared to adults. In a study by CSSCD, incidence of cerebral infarct per 100 patient years was 0.7 between ages 2-5 years while it was 0.04 between ages 20-29 years. The recurrent strokes occur in about two-thirds of SCD patient within two years of first stroke. The recurrent strokes are more common in children as compared to adults and are associated with more neurological deficits. The risk factors for cerebral ischemia includes low steady state hemoglobin, previous history of transient ischemic attack, history of acute chest syndrome and high systolic blood pressure.(30).

The role of Trans cranial Doppler (TCD) in predicting the risk for stroke in children has been well established. It measures the mean velocity of blood flow in large intracranial arteries in circle of Willis which is inversely proportional to the diameter of the vessel. In children, a mean TCD velocity >200 cm/sec in the middle cerebral artery or internal carotid artery are highly associated with increased risk of stroke. In a study by Adam R et al, 283 TCD were performed in 190 children and young adults with SCD (age at entry was 3 to 18 years). Of 190 patients, 23 patients (12 percent) had an abnormal TCD (based upon highest blood flow velocity in the middle cerebral artery), and total of seven patients developed a cerebral infarction after an average follow-up of 29 months. Among seven strokes, six occurred from the 23 patients with abnormal ultrasound results. (32,33)

In addition to infarcts, one third of SCD patients have intracranial hemorrhages which can manifest as intracerebral, intraventricular or subarachnoid hemorrhage. In contrast to cerebral infarction, which is more common in children, the peak incidence of intracranial hemorrhage is between 20-29years. About 3% of patients with SCD will have an ICH by 20 years of age, of

which 25 to 50 percent will die within two weeks of the event. The subarachnoid hemorrhage is the most common site and is result of rupture of arterial aneurysm. Multiple aneurysms are present in about 45% of SCD patients which are thought to result due to high flow conditions in cerebral circulation. The risk factors for an ICH in SCD patients include low steady state level of hemoglobin and high leucocyte count. (30,34)

In addition to overt clinical events like stroke, subtle neurological dysfunction is being increasingly recognized in patients of SCD. In a study by Vichinsky EP, neurocognitive measurements were done in neurologically asymptomatic SCD patients. In this study, as compared to healthy age matched controls, SCD patients had poorer cognitive performance.(35)

Seizure disorder is two to three times more common in SCD patients than in non-sickle populations and is associated with increased mortality. In a study by Ali SB et al, all records of the 543 persons in the Jamaica sickle cell cohort were analyzed and it was found that five-year cumulative incidence of febrile convulsions was 2.2 percent and the incidence rate of epilepsy was 100 per 100,000 person-years. The risk factors for epilepsy included male gender and history of dactylitis in childhood.(36)

3.Pulmonary Complications and Acute chest Syndrome

The slow flow and low oxygen tension in pulmonary arterial circulation forms an ideal environment for HbS polymerization and subsequent vasoocclusion. This leads to both acute and chronic pulmonary complications in SCD patients and is leading cause of mortality in SCD patients. The evaluation of SCD patients reveals variety of chronic complications like restrictive and obstructive lung disease, asthma or recurrent wheezing, pulmonary hypertension and pulmonary fibrosis.(37,38)

The baseline oxygen saturation in asymptomatic SCD patients has been found to be low as compared to healthy controls when measured by pulse oximetry. In a study by Uong EC et al, studied 130 patients of SCD and found that mean day time oxygen saturation was below normal levels.(39).

Pulmonary hypertension (PH) is increasingly being recognized as one of the major complications of SCD patients and is associated with increased mortality. The major risk factor for pulmonary hypertension is the severity of hemolysis in SCD patients. The prevalence of pulmonary hypertension is 30-40 percent in homozygous sickle cell anemia in studies where SCD patients were screened by echocardiography. However, using right heart catheterization, the prevalence of pulmonary hypertension was found to be 6-10 percent.(40,41)

The patient usually presents with exertional dyspnea and excessive fatigue. The other symptoms which may be present include pedal edema and signs of right heart failure due to right heart failure in advanced cases. The diagnosis can be confirmed by right heart catheterization which documents a mean resting pulmonary arterial pressure of >25mmHg. However the hemodynamic features may vary across the SCD patients with some having predominantly pre capillary PH while others as post capillary PH and still others may have combination of both.

The venous thromboembolism and pulmonary arterial thrombosis are common in patients of sickle cell disease. In a cross sectional study by Naik RP et al, among 404 patients, 25% had history of venous thromboembolism of which 19% were non-catheter related and the median age at diagnosis was 29.9 years.(42) In a study by Stein PD et al, data from National Hospital Discharge Survey was analyzed and it was observed that prevalence of pulmonary embolism in SCD patients was 0.44% as compared to 0.12% in patients without SCD. In addition, in patients with SCD, the pulmonary embolism was associated with longer hospital stay, greater severity of

illness as well increased mortality as compared to non-SCD patients with pulmonary embolism.(43)

Pulmonary fibrosis may is rarely seen patients with SCD and may be the result of recurrent episodes of acute chest syndrome with pulmonary infarction.(41)

Acute Chest Syndrome (ACS)

Acute chest syndrome, defined as the new appearance on an infiltrate on chest x-ray along with fever and/or pulmonary symptoms is the most common acute pulmonary complication in patients with sickle cell disease, occurring in 30-50% of patients. It is more common in children with an incidence rate of 25.3 per 100 patient years as compared to adults with an incidence rate of 8.78 per 100 patient years. It is the leading cause of mortality in adult patients with SCD with a death rate of 1.8% in children and 4.3% in adults. It is more common in homozygous sickle cell disease as compared to compound HbS/beta thalassemia with incidence rates of 3.27 and 1.95 per 100 patient years respectively. In patients of SCD, ACS is the second most common cause of hospitalization.(38)

The clinical features of ACS included fever, chest pain, pain in extremities and dyspnea, and the symptoms were more prevalent in adults as compared to children. In addition, the adult patients with SCD have prolonged hospital stay, more severe illness and increased risk of death as compared with children with SCD during an episode of ACS. ACS developed in about half of the SCD patient during the hospitalization for other complication of SCD most commonly with acute painful episode.(44)

The etiology of ACS is multifactorial and includes pulmonary infarction, infection (which can be due to viruses, Chlamydia or mycoplasma) and fat embolism. In a study by Vichensky EP, 671 episodes of ACS were analyzed and the definitive etiology could be established in 38% of ACS

episodes after extensive evaluation. It included infections (29 percent of all episodes), pulmonary infarction (16 percent) and fat embolism (9 percent). In 46% of episodes either data was incomplete or no diagnosis was established.(44) The risk factors for ACS include VOC , postoperative status, history of asthma and chronic hypoxemia.(38)

The diagnosis of ACS is made clinically on the basis of new pulmonary infiltrate which involves at least one complete lung segment detected on chest x-ray along with fever and/or pulmonary symptoms. The investigations include chest x-ray, electrocardiogram, and complete blood count along with blood and sputum cultures. In addition, CT scan of chest and bronchoscopy with bronchioalveolar lavage may be done in certain situations.

The therapy of ACS includes oxygen, analgesia for pain control, antibiotics and blood transfusion. In addition exchange transfusion may be required in some cases. In view of bronchial hyper reactivity, broncho dilators are often used.(45)

4. Osteonecrosis.

The avascular necrosis (AVN) also known as osteonecrosis is well described complication of SCD. The commonly involved sites include head of femoral and head of hemurus. The lesser common sites of involvement include condyles of mandible, elbow and temporomandibular joint.

The femoral head is the most common site of involvement. In a study of 2590 SCD patients in the Cooperative Study of SCD with a mean follow up of 5.6 years, 10% patients had AVN at entry. The overall incidence estimated was 2-4.5 per 100 patient years. The presence of alpha thalassemia mutation was associated with higher incidence of AVN while increased levels of HbF were protective.(46)

The AVN in femoral head presents with pain in the hips and progresses to decreased mobility, abnormal gait and limb length discrepancy. It is found in all ages including children as young as five years. In a study by Hernigou P et al, 95 affected hips in 52 SCD patients, who were diagnosed with AVN of the femoral heads in childhood, were studied after an average duration of 19 years after the onset of AVN, pain was present in 80 percent of the affected hips and showed permanent damage ,the pain was first noticed at an mean age of 12 years (range 7 to 15 years), and about 16%(15 of the 95) of affected hips required a surgical procedure at a mean of 30 years (range 18 to 32 years) after onset of AVN. (47)

The diagnosis is made by radiological imaging with an MRI. Although AVN is usually associated with pain, about 50% of SCD patients had AVN on imaging in surveillance study out of which 21% of patients became symptomatic subsequently.(46).

The treatment of femoral AVN includes conservative management with pain relief, walking aids and physical therapy to strengthen the hip muscles and maintain range of motion at the hip joint, and surgical interventions like core decompression and arthroplasty.

5.Infection

Infections are major contributors of morbidity and mortality in patients of sickle cell disease. As SCD patients have hyposplenism due to sickling of red cells within splenic circulation, SCD patients are susceptible to overwhelming infection by various pathogens especially by capsulated organisms like Streptococcus pneumoniae and Haemophilus influenzae. The viral infections like Parvo viruses and H1N1 can lead severe illness in SCD patients. In addition, malaria is known cause of increased mortality in children with SCD. Infections also provoke a cascade of SCD-specific pathophysiological processes which can lead various complications like VOC and acute chest syndrome. Historically, infection has been a major cause of mortality in SCD, especially in

children, and was implicated in 20-50% of deaths in various prospective cohort studies over last 20 years. (48,49).

The Streptococci pneumoniae is most common cause of bacteremia in children with SCD. Before the prophylactic penicillin and pneumococcal vaccination were regularly used in children with SCD, bacteremia due to S. pneumoniae in infants with SCD was 4-10 episodes per 100 patient years . In addition, pneumococcal bacteremia is associated with a high risk of meningitis. Although the introduction of the pneumococcal conjugate vaccine has led to significant reduction of above 90 percent in the incidence of pneumococcal infection in children below the age of five years, the infection with pneumococcal serotypes that are not included in these vaccines, and infection in those who are not vaccinated has continues to occur.(50)

The second most common organism for bacteremia in children with SCD is Haemophilus influenzae type b and is responsible for 10- 25 percent of episodes. It is more common in older children and usually less fulminant as compared pneumococcal bacteremia. The incidence of invasive H. influenzae type b infection has markedly declined after the introduction of the H. influenzae type b vaccination.(49,52)

The bacteremic episodes can lead to meningitis, pneumonia or osteomyelitis. The meningitis is especially common in young children and infants with SCD having S.pneumonea infection. Historically, as much as 50% of patients with bacteremia can develop meningitis, emphasizing the need for rapid antibiotic administration. (49)

The incidence of bacterial pneumonia in patients with SCD is high and commonly involved organisms include mycoplasma, Chylmadia and Legionella while S. pneumoniae and H. influenza type b are uncommon. In addition, respiratory viruses can also cause pulmonary

infection. Patients usually present with dyspnea, cough, fever, chest pain and tachypnea. Pulmonary infection can lead to development of acute chest syndrome.(52)

The osteomyelitis and septic arthritis are well recognized complications in patients with SCD. The most common organism involved in osteomyelitis is salmonella while staph aureus is more commonly involved in septic arthritis. The clinical features include fever, local warmth and tenderness which may be difficult to differentiate from VOC. However, infectious episodes are usually more prolonged and pain is usually localized to a single site. It may be difficult to differentiate the osteomyelitis from VOC as imaging features in both are similar. In such cases biopsy of the lesion is the gold standard for diagnosis.(53)

6. Splenic Sequestration

Acute splenic sequestration is one of the life threatening complications in SCD patients which occur when vaso-occlusion occurs within the splenic circulation that prevents blood from leaving the spleen. This leads to pooling of blood in the spleen with acute fall in hemoglobin. It is more common in children with an incidence of 30 percent and is the initial symptom in about 20 percent of patients. It is associated with 10- 15% mortality and is recurrent is 50% 0f cases. (54).

The clinical features in splenic sequestration include precipitous fall in hemoglobin concentration with reticulocytosis, tender splenomegaly, fever, leukocytosis and thrombocytopenia. It leads to decrease in effective circulating blood volume, which can lead to hemodynamic instability and shock. In rare cases splenic rupture and death may occur.

The treatment includes the blood transfusion and supportive care with pain relief, intravenous fluids and oxygen supplementation. In recurrent cases, splenectomy can be done, however, the frequency of VOC post splenectomy has been found to increase.(54,55)

7.Aplastic Crisis

The infection with Parvo virus B19 causes a serious complication in patients of SCD termed aas aplastic crisis. It is characterized by transient arrest of erythropoesis leading to abrupt fall in hemoglobin with decreased reticulocyte count. In a study by Sergeant BE, 280 patients of SCD were studied; aplastic crisis occurred in 118 of 177 parvo virus B19-infected patients. About 70% of patients had seroconvert by 20 years making parvovirus an uncommon cause of aplastic crisis in SCD after this age.(56).

The management includes transfusion therapy to maintain normal hemoglobin concentration. It is usually transient phenomenon and resolves in 10-14 days.

8. Priapism

Priapism defined as penile erection in the absence of sexual activity or desire lasting more than two to four hours, is a well-recognized complication in SCD patients. It requires prompt recognition and treatment as recurrent episodes can lead to erectile dysfunction. The prevalence among boys and men with SCD ranges from 6 to 45 percent. The episodes of priapism can occur at any age, but is usually a more significant clinical problem after puberty. The mean age of onset is 12 - 15 years; with 75- 90% patients report their first episode prior to 20 years of age. (57)

The patients with can present with either as prolonged erection if it lasts for more > 3 hours or as stuttering episodes if it lasts for a few minutes but < 3 hours and resolves spontaneously or combination of both.(58)

Although most episodes may resolve spontaneously, prolonged episodes need urgent medical attention with aspiration and irrigation of corpus cavernosum.(57,58)

9. Hepatobiliary Complications

The liver is affected by a variety of SCD complication both directly as well as due to treatment of SCD. The various manifestations include acute hepatic sequestration crisis, benign cholestasis and sickle cell hepatopathy. The complications due to treatment of SCD include liver involvement due to transfusional iron overload, viral infections like Hepatitis B and heaptitis C due to transfusion related transmission. (59)

Cholilithiasis is common in patients with SCD as chronic hemolysis leads to formation of pigment stones and is found in about 70 percent of patients. Choliltiasis can be asymptomatic or can present with acute cholecystitis, chronic cholecystitis or with choledocholithiasis. In a study by Adam S eta al, 509 adult patients with SCD were studied; cholecystectomy was done in 48% of patients between 18-47 years of age and in 69% of patients aged above 47 years. (59, 60)

10.Growth and Development

In children and adolescents, SCD is associated with delayed sexual maturation, growth retardation and being under weight. In a study by Rhodes et al, 33 SCD adolescent patients along with age matched controls were studied for growth parameters. They demonstrated that growth is delayed during puberty in the adolescent patients with SCD and this delay is independently associated with decreased hemoglobin concentration and increased total energy expenditure. The normal height is often attained by adulthood but weight usually remains lower than that of controls. Both neurodevelopment and skeletal maturation are usually delayed.(61).

In Jamaican Cohort Study by Singhal A et al, extreme growth retardation which was defined as absence of both the adolescent growth spurt as well as prepubertal sexual development (Tanner stage 1 or 2) at the age 16, was found in 8 of 52 boys (15%) with SCD.(62)

11.others

The SCD is a multisystem disease and any system can be affected by the disease process. In addition, SCD is known for considerable phenotypic heterogeneity in presentation. The other common complications which can occur in SCD patients include leg ulcers, retinopathy and nephropathy.

Leg ulcers are more common in homozygous sickle cell anemia as compared to compound heterozygous states like sickle beta thalassemia and occur in about 10 per 100 patient years of SCD. In a study of 225 SCD patients from the Jamaican Cohort Study, 53 patients (24 percent) had chronic leg ulcers and median age of first ulceration was about 17 years (range: 13 to 24) . The mode of onset was traumatic in 60% while it occurred spontaneously 28%. The risk factors detected were venous incompetence, increased levels of LDH and low socioeconomic status. The medial and lateral maleoli are most common site of involvement.(63)

The ocular manifestation of SCD is one of important long term complications of SCD that have emerged because of the increase in life expectancy of SCD patients. SCD can affect any ocular structure but retina is most commonly involved and leads to proliferative retinopathy. It is more common in HbSC disease as compared to homozygous sickle cell anemia. Although spontaneous regression of retinopathy is known, loss of vision can occur in untreated cases. (64)

The kidney involvement occurs in 30-50% of adult patients with SCD and leads to renal failure in 18% of patients. It can manifest as asymptomatic albuminuria or patient may present with enuresis due to hyposthenuria, hematuria, and proteinuria with hypertension or with renal colic due to papillary infarction. The rarer presentation include nephrotic syndrome and nephrogenic diabetes insipidus.(65)

V. Phenotypic Heterogeneity:

The clinical features of SCD are exceptionally heterogeneous. Clinical heterogeneity is present among the different genotypes of sickle cell disease (eg, SCA, HbSC disease, HbS-beta thalassemia) as well as in homozygous HbS. Although identical beta globulin gene mutation is present in all patients, the clinical course of patients with homozygous sickle cell anemia is highly variable. This suggests that product of other genes known as epistatic genes along with environmental factors are likely to determine the specific phenotype of SCA. (66)

The various modifiers of clinical expression are described below:

1.Fetal Hemoglobin (HbF):

Fetal hemoglobin (HbF) is the most thoroughly studied as well as most powerful genetic modulator of sickle cell disease. The role HbF in SCD was observed long back when it was noticed that infants with SCD do not have symptomatic disease and it was attributed to high HbF levels present during early infancy. The increased HbF levels are associated with a decreased rate of acute pain episodes, less osteonecrosis, fewer leg ulcers, and lesser frequency of acute chest syndromes as well as reduced disease severity. However, the level of HbF do not show a clear association with complications like priapism, proteinuria, stroke and silent cerebral infarction, systemic hypertension, and pulmonary hypertension. The HbF level in SCA varies from 2-8% but varies with various beta gene haplotype.(67)

The two main properties of HbF which are considered to ameliorate SCD symptoms include:

1. HbF molecules do not participate in polymerization that occurs between molecules of deoxyHbS which results in decreased polymerization and in turn milder clinical phenotype.
2. The higher concentration of HbF in a cell infer lower concentrations of HbS which in turn decreases polymerization.(66)

The various factors that affect the level of HbF include the haplotype of beta gloobulin gene cluster, polymorphism at various quantitative trait loci (QTL) such as HBS1L-MYB intergenic region (HMIP) region and BCL11A (2p16). (67)

2.Alpha Thalasesemia:

Concurrent alpha thalassemia has been found in approximately 30% of patients with SCD and is considered a modifier of sickle cell disease severity. SCD patients who have coincidental alpha thalassemia have lesser hemolysis and overall milder clinical phenotype. Alpha-thalassemia decreases mean corpuscular volume, increases hematocrit and reduce the concentration of HbS and HbS Polymerization. Vaso-occlusive events that are highly dependent on hematocrit, such as stroke, leg ulcer and splenic function, appear to benefit from the coexistence of alpha-thalassemia. In contrast, adult patients with SCA with alpha thalassemia usually do not have a reduction and may have an increase in certain complications like painful episodes, acute chest syndrome and avascular necrosis, and is attributed to increased blood viscosity as a result of higher hematocrit resulting from associated alpha thalassemia mutation.(68)

3.Hemoglobin Haplotype of Beta globulin (HBB gene like cluster):

The other potential modulator of sickle cell disease phenotype is haplotype of beta globulin gene.

The HbS beta globulin gene is found in five common haplotypes, described as polymorphic restriction endonuclease sites in and around the mutant ß-globin gene. Although haplotypes have numeric identifiers, they are most commonly designated by the geographic areas in which they were first identified: Senegal, Benin, Central African Republic (or Bantu), Cameroon and Saudi-Indian (or Asian). Carriers of the HbS gene on Senegal or Saudi-India haplotype usually have the highest HbF level and PCV and the mildest clinical course. Individuals with Bantu (Central African Republic) haplotypes have the lowest HbF level and PCV and the most severe clinical course. The carriers of all haplotypes have considerable variation in HbF levels, which suggests the importance of other quantitative trait loci (QTL) in modulating beta globulin gene expression.(67)

4.Genetic Polymorphism

Neither HbF level nor beta globin genotype can completely explain the clinical and laboratory diversity of SCD. Both HbF and alpha thalassemia affect the phenotype of disease by decreasing the polymerization of HbS. In a study of 1265 SCD patients with either "severe" or "mild" disease based on a defined model of disease severity, a genome-wide association study (GWAS) found 40 single nucleotide polymorphisms (SNPs) were strongly associated with sickle SCD severity (with an odds for association >1000). Of these, 32 SNPs were further analyzed in a separate set of 163 SCD patients, five were replicated, eight showed consistent effects but failed to reach statistical significance and 19 did not show any conclusive association.(69)

The polymorphic genes that are thought to be important in phenotypic variability of SCD include genes involved in TGF-β/BMP pathway.(66)

VI. Management of SCD and Role of Hydroxyurea:

The management of SCD includes diagnosis, prevention and treatment of its complications and potential cure of this illness and is briefly described below.

1.Diagnosis

The diagnosis of SCD essentially consists of analysis of hemoglobin which can be done by protein electrophoresis or by chromatography. The various diagnostic tests include peripheral smear review, sickle solubility test, Hb electrophoresis, High Performance Liquid Chromatography (HPLC) and DNA based tests to detect the specific mutation. The type of test used depends upon the age of the patient as well as clinical scenario. DNA based tests are used in prenatal diagnosis and in patient with recent history of blood transfusion, while HPLC in combination peripheral smear review and sickle solubility test is used in most of the other patients. The neonatal screening programmes are in place in certain countries like US and England and leads to early diagnosis of SCD. The electrophoresis at alkaline pH (cellulose acetate electrophoresis) or at acidic pH (Citrate agar electrophoresis) combined with sickle solubility test allows definitive diagnosis of SCD. (70).

The HPLC is highly precise and automated technique for identification and quantification of hemoglobin. This technique detects most hemoglobin variants by their different retention times. It is highly sensitive as well as specific and provides both the quantitative and qualitative interpretation .(70)

The measurement of HbA2 is valuable for diagnosis of compound heterozygous sickle beta thalassemia as it is increased in such cases. However, it should be noted that HbA2 may be

overestimated in presence of HbS and in some cases familial studies and/or DNA based test may be required for the confirmation of diagnosis.(70)

2.General Overview and Prevention of Complications:

The management of SCD requires needs comprehensive care, team management along with education of the patients and their family members. It needs careful monitoring of growth and development in children and needs early detection of complications. The various primary preventive measures includes routine health management, use of penicillin prophylaxis, immunizations, and regular blood transfusions for certain patients. The only US Food and Drug Administration (FDA) approved therapy for prevention of various complications of SCD is the use of hydroxyurea which reduces sickle hemoglobin polymerization process by increasing the production of HbF levels .(71)

The penicillin prophylaxis should be started in all children at diagnosis and continued till at least till five years of age as children are at high risk of sepsis especially invasive pneumococcal disease due to functional asplenia.(71)

In 2012 Cochrane review by Riddington C et al, data from three such trials (457 patients of SCD) was analyzed . As compared with placebo or no treatment, the penicillin prophylaxis was associated with a reduced risk of pneumococcal infection (odds ratio 0.37; 95% CI 0.16-0.86) as well as decreased risk of death (odds ratio 0.11; 95% CI 0.01-2.11). (72)

The immunization forms the cornerstone of infection prevention in SCD patients. Children with sickle cell disease should receive all routine vaccines especially against pneumococcus, Meningococcus, Haemophilus influenzae and hepatitis B. (71). The use pneumococcal

vaccination has led to significant decrease in incidence of invasive pneumococcal disease in children with SCD. (50)

3. Blood Transfusion and Chelation Therapy:

Blood transfusion is used both for prevention as well as treatment of SCD related complications. The phenotype may vary transfusion dependent anemia requiring regular blood transfusions to chronic anemia with occasional acute drop in hemoglobin requiring blood transfusion. The role regular blood transfusion in prevention of certain complications like stroke in children is well established. However, blood transfusion carries the risks as well and the risk benefit ratio should be carefully assessed in these situations.(73)

The prophylactic blood transfusion has been advocated in primary and secondary prevention of stroke, silent cerebral infarcts, and preoperative management and in patients with chronic anemia requiring regular transfusions. (73).

In a study by Adam RJ et al, 130 children with sickle cell anemia and Trans Cranial Doppler (TCD) velocity of more than 200cm/sec on two occasions with no prior history of stoke were randomly assigned to institution of prophylactic transfusion to decrease HbS concentration below 30% or regular standard care. The study was prematurely terminated after 20 months of follow because of marked benefit to prophylactic transfusion group(10 cerebral infarctions and 1 intracerebral hemorrhage in the control group, as compared to1 infarction in the transfusion group ($P<0.001$)). (74)

The blood transfusion may be given for treatment of various complications of SCD and includes acute splenic sequestration, aplastic crisis, acute chest syndrome and acute symptomatic anemia .

The other less frequent indications of chronic blood transfusion therapy in SCD patients include recurrent ACS despite on hydroxyurea, severe recurrent acute painful crisis not responding to hydroxyurea, recurrent priapism and third trimester of pregnancy.(73)

In SCD patients requiring blood transfusion for decreasing the level of HbS, simple blood transfusion may not be appropriate as it will be associated with increased blood viscosity and may not decrease the HbS to desired level. In such cases, exchange transfusion is done which involves removing of patients' blood and then replacing it with exogenous blood. Exchange transfusion has been found to be more beneficial in case of stroke, respiratory failure and multi organ failure.(75)

The initiation of chelation therapy in SCD patient dependents upon the number of transfusions given, the degree of hepatic and cardiac iron deposition in the amount of their dysfunction present, and the type of transfusion regime. The chelation therapy is recommended when Serum Ferritin is more than 1000 microgram/L , Liver Iron Concentration is more than 7 mg/g dry weight and cumulative transfusions of more than 120 cc of packed RBCs/kg .(76)

4. Pain management:

The acute painful episodes are the most common cause for patients with SCD to seek medical attention. The treatment consists of aggressive pain relief including the use of opiates, other analgesics, or even other modalities. In addition, exclusion of causes other than vasoocclusion (particularly infection), optimal hydration by oral or intravenous fluid resuscitation (especially in children) is also important. Blood transfusion may be required in patients having hemodynamic instability and there is no role of blood transfusion to decrease the severity of pain. The acute

pain episodes can be managed at home or hospital depending upon the severity. In hospital parentral opiod analgesia should be started and dose titrated as per the response.(77)

5. Cure:

A life-long cure for SCD is available only through hematopoietic stem cell transplantation (HSCT) and its use in SCD is evolving. However, hematopoietic stem cell transplant has been primarily used in children with severe phenotype where HLA identical sibling has been available; there is scarcity of data regarding the use transplant with alternative donor or adult patients with SCD.

Historically only myeloablative conditioning regimens have used for HSCT which were associated increased complications especially in adults. However ,in a study by Mathew M eta al, 30 patients with SCD were given non myeloablative conditioning with Alemtuzumab, total body irradation (300cGY) and sorafenib, after median follow up of 3.4 years, 87% of patients had long term stable donor engraftment without acute or chronic graft versus host disease.(78)

Role of Hydroxyurea:

The hydroxyurea, first synthesized by Dressler and Stein in 1869 in Germany, is a small molecule that inhibits DNA synthesis by blocking to ribonucleotide reductase. This drug was first tested for solid organ malignancy and in subsequent decades, it was used for multitude of diseases including chronic myeloid leukemia, psoriasis, ovarian carcinoma and polycythemia vera.(79)

In 1984 hydroxyurea was first time in adult patients with SCD as it was found to increase HbF levels and subsequently in 1998, FDA approved the use of hydroxyurea in symptomatic adult sickle cell patients. The increased HbF concentration within RBCs results in less polymerization of abnormal hemoglobin.(79)

In landmark study, The Multi Centre Study of Hydroxyurea in Sickle Cell Anaemia (MSH) by Charache et al, 299 adult symptomatic sickle cell patients(defined as three or more painful crisis per year) were evaluated in a randomized controlled trial. 152 patients were assigned to Hydroxyurea (HU group) while 147 were assigned to placebo group and were followed for an average of 21 months. . The hydroxyurea use was associated with significant reduction in painful crisis(4.5 per year to 2.5 per year) along with marked reduction in incidence of acute chest syndrome (25 vs. 51, $P<0.001$) and blood transfusions(48 vs. 73, $P< 0.001$). The starting dose of hydroxyurea was 15mg/kg up to a maximum tolerated dose of 35mg/kg. There were no significant side effects of hydroxyurea use in this trial. (80) The 9-year follow up of same study revealed 40% reduction in the mortality among those who received hydroxyurea. The survival was associated with HbF level and frequency of painful episodes.(81)

After the successful use of hydroxyurea in adults with severe symptomatic SCD, its use in pediatric patients was explored. In a study by Ferster A, 25 children with SCD with a mean age of 9 years were randomized to treatment with 15mg/kg of hydroxyurea for six months or placebo and then switched to other arm for six months. Out of 22 evaluable patients, 16 showed complete resolution of symptoms requiring hospitalization during treatment period(73% reduction).(82)

In a pilot study by Winfred C et al, 28 children with SCD with a median age of 15 months were administered 20mg/kg/day of hydroxyurea to assess the feasibility and use in infant population.

After the two years of follow up, authors concluded that hydroxyurea use is feasible in infants and may delay organ dysfunction. The subsequent follow up of same study after four years revealed decreased incidence of acute chest syndrome(7.5 versus 24.5 in historical controls per 100 patient years),better splenic function and improved growth. A further follow up of same cohort after about mean follow up of fifteen years have sustained clinical and hematological benefit of hydroxyurea with no significant side effects.(83,84,85)

In a study by Winfred C et al, 193 children aged between 9-18 months with SCD were randomized (98 to hydroxyurea group and 97 to placebo) to either receive hydroxyurea 20mg/kg/d or placebo for a period of two years. the primary end points were assess organ dysfunction, clinical complications and toxicity of treatment. After the mean follow up of two years, there was no significant differences between the groups for the organ dysfunction (19 out of 70 had decreased spleen function in the hydroxyurea group versus 28 out of 74 patients in the placebo group, p=0·21; and the difference in the average increase in DTPA glomerular filtration rate in the hydroxyurea group versus the placebo group was 2 mL/min per 1·73 m^2, p=0·84). However, use of hydroxyurea significantly decreased acute painful episodes (177 events in 62 patients in HU group versus 375 events in 75 patients in placebo group, p=0·002) and dactylitis (24 events in 14 patients versus 123 events in 42 patients in placebo group, p<0·0001), with some evidence for decreased frequency of acute chest syndrome, hospitalization rates as well as transfusion. Hydroxyurea use increased hemoglobin and fetal hemoglobin, and decreased leucocyte count. The toxicity noted was limited to mild-to-moderate neutropenia.(86)

The current guidelines recommend the use of hydroxyurea in adults with 3 or more severe vasoocclusive crises per year, chronic pain or chronic anemia interfering with activities of daily living , or patients having severe or recurrent episodes of acute chest syndrome. In children

treatment with hydroxyurea is suggested in symptomatic older children as well as in asymptomatic infants (after the age of 9 months). (71, 87)

Aims & Objectives

Aims and objectives:

1. To analyze the demographic, clinical and laboratory profile of patients (adults and children) with sickle cell disease.

2. To assess the use and response to hydroxyurea in these patients.

3. To identify the demographic, clinical, and laboratory parameters that can

predict response to hydroxyurea in these patients.

Patients & Methods

Patients and Methods:

This study protocol was approved by our Institutional Review Board (IRB).

This study involved a retrospective analysis of patients with sickle cell disease from 2005-2013 who were treated in Department of Clinical Haematology

Duration of the study: June 2014 to December 2014.

Settings of the study: Department of Clinical Haematology.

Diagnostic Criteria:

Sickle cell disease was diagnosed in patients by either of the following methods:

1) Hemoglobin variant analysis done by High performance Liquid Chromatography (HPLC) showing either homozygosity for HbS or HbS beta thalassemia.(70)
2) Hemoglobin mutation analysis done by reverse dot blot technique revealing either homozygous HbS or HbS beta thalasemia.(70)

Inclusion criteria:

1. All the patients seen and diagnosed as having sickle cell disease (homozygous sickle cell and sickle beta thalassemia) and have had atleast six months of subsequent follow up were included in the study.

Exclusion criteria:

1. Patients with other hemoglobinopathies.
2. Patients with follow up duration < 6 months or no follow up or where the data was not retrievable.

Collection of data:

After approval by the Institution Review Board, the patient data base at our institution was reviewed to identify all patients (adults and children) diagnosed and treated for sickle cell disease at our institute from January 2005 to December 2014. Medical information regarding the clinical/laboratory details at diagnosis, post treatment response and adverse events were obtained from the hospital records (laboratory reports/ physician documentation in hospital charts/hospital discharge summaries).

Only patients who had at least six months follow up after starting treatment were categorized as 'evaluable' for assessment of response.

Treatment: All patients who were diagnosed with sickle cell disease and had adequate follow up were included in the study. Data was collected with regard to demographic ,clinical and laboratory profile of the patients, use of hydroxyurea , assessment of its response as well as adverse events were noted. The dose of hydroxyurea varied from 10mg/kg/day to 35mg/kg/day

Data analysis:

Results are analyzed in terms of the demographic, clinical characteristics and laboratory parameters at diagnosis, and at different time interval after starting of Hydroxyurea therapy. The response to treatment is assessed in terms of Complete Response (CR), Partial response (PR) and No response. The CR in sickle cell disease was defined as complete resolution of presenting symptom while partial response was defined as at least 50% reduction in frequency of presenting symptoms. The diagnosis of anemia was done as per the WHO definition. (88)

The following parameters were analyzed

a) Demographic, clinical, and laboratory profile at diagnosis
b) Frequency of presenting symptom and changes in laboratory parameters like HbF after three and six months of hydroxyurea treatment.
c) Frequency of symptoms at last follow up
d) Dose, compliance, response and adverse events to hydroxyurea therapy

All patients with a minimum follow up of six month were considered evaluable for response . The closing date for analysis was March 31, 2015.

STATISTICS

Descriptive statistics were calculated for all variables. Differences in proportions and two categorical variables were assessed using the chi-square test. Mann-Whitney-U test was used to compare two continuous variables. For all tests, a 2-sided P-value of 0.05 or less was considered statistically significant. SPSS 16.0 software was used for the analysis.

Results

RESULTS

From January 2005 to December 2013, a total of 9389 patients were evaluated in department of Clinical Haematology for suspected hemoglobinopathy by hemoglobin variant analysis or thalassemia mutation analysis (or both where appropriate). Of these, 745(7.9%) were diagnosed as sickle cell disease (consisting of homozygous sickle cell disease and compound heterozygous sickle beta thalassemia). Out of this, only 230 patients had adequate follow up and were included in present study.

There were total of 230 patients of sickle cell disease who fulfilled the inclusion criteria and were included in this study for analysis. (Figure 1)

Certain data was available only on a portion of the patients. For each result therefore, the numbers of patients included are mentioned accordingly.

DEMOGRAPHY PROFILE AT DIAGNOSIS: (Table: 1)

The median age of the 230 patients was at the time of evaluation was 12 years (range: 1-54 years), while median age at which first symptom occurred was 4 years (1-54 years). Males were predominantly represented in the study group i.e. 152 (66.1%) males and 78 (33.9%) females. The male female ratio was 1.9:1. They were patients from different states of the country and two patients from outside country (Maldives). (Figure 2)

The family history of similar illness was present in 36(15.7%) patients while majority of patients (n-194, 84.3%) had no history of any such illness in family

SYMPTOMS AT DIAGNOSIS AND OVERALL FREQUENCY OF VARIOUS COMPLICATIONS: (Table No.2 and Table No.3a and Table No.3b)

The bone pain was the most common symptom and was present in 187(81.3%) and was the only symptom in 93 patients (40.4%). Anemia as defined by WHO criteria (as described above) was present in 227(98.6%) patients while symptomatic anemia was presenting symptom in 125 patients (54.5%) out of which 10 patients (4.3%) were having transfusion dependent anemia. In addition, patients presented with combination of pain and anemia (68 patients, 29.6%). The symptoms combinations at diagnosis were joint pain and avascular necrosis in 11 patients (4.8%) and pain, anemia and avascular necrosis in two patients (0.9%). History of blood transfusion was present in 167(72.6%) patients. Other clinical features which were present at diagnosis included abdominal pain in 28(12.2%), jaundice in 14(6.1%) and febrile episodes in 17(7.4%). Splenomegaly was present in 115 patients (50%) with an average size of 3.7cm (range 2-9cm) below left costal margin.

A number of complications were detected in our cohort of patients at diagnosis or during the period of follow up. This included avascular necrosis of femur head in 32 patients (13.9%), cholilithiasis in 14(6%) and acute chest syndrome in 14 (6.1%). Various infections were documented in 11 patients (4.7%) and this included osteomyelitis (n-5), septic arthritis (n-2), pneumonia (n-2) and urinary tract infections (n-2). The lesser common complications detected during follow up included chronic leg ulcer in 3 patients (1.3%), seizure disorder in 2 patients (0.8%) and stroke in 2 patients (0.8%)

LABORATORY PARAMETERS AT DIAGNOSIS (Table No.4)

Out of 230 patients, majority of the patients [n=227(98.6%)] were found to have anemia (as per WHO defined criteria). The median hemoglobin at presentation for the entire cohort was 8.7g/dl (range: 4-12.5g/dl). The white cell count was available for 229 patients and median WBC count was 11660/mm^3 (range: 2100-39600). The median corpuscular volume in 223 patients (where data was available) was 80.6fl (range: 57.7-108.2fl).

The reticulocyte count was available in 218 (94.7%) patients and median reticulocyte percent for these patients was 7.06% (range: 0.69-23.76). The median platelet count at presentation was 274448/ mm^3 (range: 11000-3449000). The percentage of Hemoglobin F and Hemoglobin S at diagnosis was available for 182 patients (79.1%). The median HbF percent was 19.5 (range: 4.7-54%) while median HbS percent at diagnosis was 72.2 (range: 28-87%).

Using a combination of variant analysis (n = 182) and RDB (n = 48), 148 patients (64.3%) were diagnosed to have homozygous sickle cell disease while 82 patients (35.6) had sickle beta thalassemia

TREATMENT AND RESPONSE (Table No.5 and Table No. 6)

Among the total 230 patients included in the study, 222 (96.5%) were treated with hydroxyurea, while 8 (3.5%) were kept under regular follow up and hydroxyurea was not started in view of minimal symptoms.

The median dose of hydroxyurea used was 15mg/kg/day (range: 10-35mg/kg/day). The response to therapy was assessed 6 months and at last follow up.

Adverse events to hydroxyurea use were noted in 17 patients (7.6%) and included leucopenia in 10 patients (4.5%), thrombocytopenia in 4 patients (1.8%) and marrow aplasia in one patient (0.4%). There were no deaths related to the use of hydroxyurea. One patient with bone marrow aplasia had full recovery of blood counts after about one year of observation.

The response assessment was done in terms of resolution of/or decrease in frequency of presenting symptoms and were defined in terms of complete response, partial response and no response (as described above).

At six months of follow, data for three patients was lacking and out of the 219 patients evaluable for response, 170 patients (77.6%) showed complete response, 43 patients (19.6%) showed partial response and there was no response in 6 patients (4%).

At last follow up, with median duration of 36.25 months (range 6-117months), 131 patients (57.4%) showed complete response while 86 patients (37.4%) showed partial response and only 5 patients (2.2%) did not show response to hydroxyurea.

EFFECT OF HYDROXYUREA ON CLINICAL AND LABORATORY PARAMETERS (Table No.7)

The comparison of clinical and laboratory parameters at diagnosis and after treatment with hydroxyurea for median duration of 36.25 months showed significant decrease in annual frequency of painful episodes from 3.4±1.3 to 0.9±0.49 (p=0.000), blood transfusion requirement from 4 units(±2.3) to 1unit(±0.25) and episodes of acute chest syndrome from 6.3% to 1.8%(p=0.01). There was significant increase in hemoglobin (g/dl) from 8.7±1.44 to 9.6±1.45 (p=0.000) and in 34 patients where pre and post treatment HbF values were available, mean HbF

(%) increased from 18.3±6.6 to 25.9±10.06 (p=0.000). The mean leucocyte count decreased from $11.71x10^3$ to $9.0±10^3$ (p=0.000)

SURGERY RELATED VARIABLES AND OUTCOME IN SCD PATIENTS (Table No.8)

There were 33 surgeries performed in 230 patients during the median follow up of 36.25 months (6-117 months). Out of these 27 surgeries were performed at CMC hospital and were further analyzed for various variables and outcome. Majority were orthopedic surgeries for avascular necrosis of femoral head and included hip arthroplasty and core decompression (16 patients - 59.2%). Other surgeries included cholecystectomy in 4(14.8) and splenectomy in 4(14.8). In addition, mitral valve replacement, hepaticojejunostomy and lower segment cesarean section were performed in 1 patient each. Most patients were managed with simple blood transfusion in perioperative period, given in 24 patients (88.8%) while 6 patients (25.9%) required exchange transfusion prior to surgery. Of the 6 patients who had exchange transfusion prior to surgery, 4 patients required perioperative blood transfusion as well (Median 1 unit; range 1-3units). Out of 27 patients, 2 patients did not require any blood transfusion or exchange transfusion. Postoperative complications occurred in 4 patients (14.8%) and included pneumonia/acute chest syndrome in one patient while other patients had complications unrelated to sickle cell disease (hematemesis in one patient, increased blood loss during surgery in one patient and urinary tract infection in one patient). Post-operative ICU care was required in 4 patients (14.8%) and there were no deaths reported.

COMPARISON OF HOMOZYGOUS SICKLE AND SICKLE BETA THALASSEMIA:

A. CLINICAL PARAMETERS (Table No.9 and Table No.10)

Out of 230 patients, 148 patients (64.3%) had homozygous sickle cell disease while 82 patients (35.6) had sickle beta thalassemia. In homozygous sickle cell anemia, 91 patients (61.5%) were males and 57 patients (38.5%) were females. In sickle beta thalassemia, 61 patients (74.4%) were males while 21(25.6%) were females.

There were no statistically significant differences in age at first symptom (4 years in both, p=0.5), however, the age of evaluation was 9 years in patients with sickle beta thalassemia as compared to 14 years in patients with homozygous sickle cell disease (p=0.03).

The most common symptom in both groups was acute painful episodes and was present in 123 patients (83.1%) with homozygous sickle and 64 patients (78%) with sickle beta thalassemia (p=0.37). There were no statistically significant differences with respect to frequency of anemia (98 versus 54, p=1.00), avascular necrosis (23 patients versus 8 patients, p=0.33), acute chest syndrome (12 patients versus 2 patients, p=0.14) and cholilithiasis (12 patients versus 2 patients, p=0.14). However, there was significantly higher incidence of splenomegaly in patients of sickle beta thalassemia as compared to homozygous sickle cell disease (37% versus 73%, p=0.00).

B. LABORATORY PARAMETERS (Table No.11)

In comparison to homozygous sickle cell anemia, the hemoglobin at diagnosis was lower in sickle beta thalassemia, but the difference was not statistically significant (9.1g/dl versus 8.6g/dl, p=0.15). The mean corpuscular volume was significantly higher in patients of homozygous sickle cell anemia as compared to patients of sickle beta thalassemia (86.6fl versus 70fl, p=0.00).

The mean WBC count was higher in patients of homozygous sickle cell anemia (11500/mm^3 versus 9550/mm^3, p=0.003) and mean reticulocyte percent was higher in patients of homozygous sickle cell anemia (6.8% versus 5.2%, p=0.001). In 182 patients (120 of homozygous sickle cell disease and 62 of sickle beta thalassemia), where data for HbF and HbS was available for both groups, patients with homozygous sickle cell anemia had higher levels of HbS (73.2% versus 70.4%, p=0.001), while HbF was similar in both groups (19.8% versus 19.2%, p=0.9).

C. USE AND RESPONSE TO HYDROXYUREA (Table No.12 and Table No.13)

Hydroxyurea was used in 145 patients (98%) with homozygous sickle cell disease whereas 77 patients (93.9%) of sickle beta thalassemia were treated with hydroxyurea but the difference was not statistically significant (p=0.137). The median dose of hydroxyurea was similar in both groups (15mg/kg/day). The adverse events occurred in 12 patients (8.2%) with homozygous sickle cell anemia and in 4 patients (6.4%) with sickle beta thalassemia (p=0.6).

Response assessment at six months of hydroxyurea therapy was available in 142 patients with homozygous sickle cell anemia (data was inadequate in 3 patients and were excluded from analysis and 77 patients with sickle beta thalassemia. At six months of hydroxyurea therapy, a significantly higher number of patients with homozygous sickle cell disease showed complete response as compared to patients with sickle beta thalassemia [117 patients (82.4%) versus 53 patients(68.8%)] (p=0.04) . In patients with homozygous sickle cell disease, 23 patients (16.2%) showed partial response while 2 patients (1.4%) showed no response at six months assessment of hydroxyurea use. In patients with sickle beta thalassemia, 20 patients (26%) showed partial response and 4 patients (5.2%) showed no response to hydroxyurea.

Long term response assessment was done at a median duration of 36.25 months (Range: 6 - 117). 88 patients (60.6%) with homozygous sickle cell disease showed complete response, 55 (37.9%) showed partial response while 2(1.4%) showed no response to hydroxyurea therapy. In patients with sickle beta thalassemia, 43 patients (55.8%) showed complete response, 31 patients (40.3%) showed partial response while 3 patients (3.6%) did not show any response to hydroxyurea therapy. The difference in response between the two groups was not statistically significant. (p=0.4)

FACTORS PREDICTING RESPONSE TO HYDROXYUREA TREATMENT (Table No. 14 and Table No.15)

In the present study, all patients who showed response (either PR or CR) were considered as responders even if they were subsequently lost to follow up. The response was defined as complete response (CR) if there was complete resolution of presenting symptom after hydroxyurea therapy, partial response (PR) if there was at least 50% reduction in presenting symptom and those who did not show response till last follow up were considered as non-responders (NR). A detailed analysis was done of factors affecting the complete response. However a significant association was observed with higher hemoglobin (p= 0.03), low reticulocyte percent (p=0.03) and higher HbF percent (p=0.01) at diagnosis. In addition, increased level of HbF at last follow up were associated with complete response (p=0.004) No significant association was found with age at presenting symptom (p=0.143), gender (p=0.112), homozygous sickle cell anemia versus sickle beta thalassemia (p=0.546), presenting symptom at diagnosis (p=0.154), leucocyte count (p=0.06), or platelet count (p=0.140) at diagnosis with the response status.

Tables and Figures

Table No.1: Demographic Profile of Patients (n-230)

Variable	N(%)/median(range)
Age at evaluation(years)	12(1-54)
Age at first Symptom	4(1-54)
Gender	
Male	152 (66.1)
Female	78 (33.9)
Family History	
Present	36(15.7)
Absent	194(84.3)
State	
Andhra Pradesh	44(19.1)
Bihar	2(0.9)
Orissa	23(10)
Jharkhand	72(31.3)
Chhattisgarh	14(6.1)
Tamil Nadu	36(15.7)
West Bengal	23(10)
Others*	16(7)

*Includes two patients from Maldives.

Table No.2: Symptoms at Diagnosis (n-229)*

Variable	N (%)
Pain	93(40.4)
Symptoms of Anemia Transfusion dependent anemia	55(20.9) 10(4.3)
Pain and Avascular necrosis(AVN)	11(4.8)
Pain and anemia	68(29.6)
Pain, AVN and anemia	2(0.9)

*One case was asymptomatic and was diagnosed screening in view of family history of sickle cell disease.

Table no.3a Overall frequency of clinical features (n-230)

Variable	N (%)
acute painful episodes	187(81.3)
Anemia*	227(98.6)
Blood Transfusions	167(72.6)
Abdominal Pain	28(12.2)
Febrile episodes	17(7.4)
Jaundice	14(6.1)
Splenomegaly	115(50)

*As per WHO defined criteria

Table no.3b Overall frequency of Complications (during follow up)(n-230)

Variable	N (%)
Avascular necrosis	32(13.9)
Acute chest syndrome	14(6.1)
Cholilithiasis	14(6)
Infections	11(4.7)
Osteomyelitis	5(2.1)
Septic arthritis	2(0.9)
Pneumonia	2(0.9)
Urinary Tract Infections	2(0.9)
Leg Ulcer	3(1.3)
Seizure disorder	2(0.9)
Stroke	2(0.9)

Table No. 4 Laboratory Parameters at Diagnosis

Variable	Mean(range)
Hemoglobin(g/dl) (n-230)	8.7(4-12.5)
Mean corpuscular volume (n-223)	80.6(57.7-108.2)
WBC count($x10^3$cells/mm^3) (n-229)	11.6(2.1-39.6)
Platelet (($x10^3$cells/mm^3) (n-228)	274(11-344)
Reticulocyte (%) (n-218)	7.06(0.69-23.76)
HbF(%) (n-182)	19.5(4.7-54)
HbS(%) (n-182)	72.2(28-87)
Creatinine(mg/dl) (n-212)	0.5(0.2-1.3)

Table no 5 Use of hydroxyurea

Variable	**N(%)/median(range)**
Hydroxyurea	
Yes	222(96.5)
No	8(3.5)
Dose(mg/kg/day)	15(10-35)
Adverse Events	17(7.6)
Leucopenia	10(4.5)
Thrombocytopenia	4(1.8)
Aplasia	1(0.4)

Table No.6 Response to Hydroxyurea

Response At 6 months(n-219)	
CR	170(77.6)
PR	43(19.6)
NR	6(2.7)
Response At last follow up(n-222)	
CR	131(57.4)
PR	86(37.4)
NR	5(2.2)

Table No.7 Effect of Hydroxyurea on clinical and laboratory parameters (n-222)

Variable	**At diagnosis Mean (±SD)/n(%)**	**After HU therapy (At last follow up) Mean (±SD)/N(%)**	**P value**
Bone and Joint pain(episodes/year)	3.4(1.3)	0.9(0.49)	**0.000**
Blood transfusion (units/year)	1.7(2.0)	1(0.25)	**0.000**
Acute chest syndrome (no. of episodes)	14(6.3)	4(1.8)	**0.01**
Hemoglobin(g/dl) (n-220)	8.7(1.44)	9.6(1.45)	**0.000**
WBC count(x 10^3 cells/mm^3) (n-220)	11.71(5.5)	9.0(3.8)	**0.000**
Mean corpuscular volume(fl) (n-215)	80.48(11.11)	88.58(15.9)	**0.000**
HbF(%) (n-34)	18.3(6.6)	25.9(10.06)	**0.000**

Table.No.8 Surgery related variables and outcome in SCD patients (n-27)

Variable	N (%)
Type of surgery	
Hip Arthroplasty	16(59.2)
Cholecystectomy	4(14.8)
Splenectomy	4(14.8)
Others*	3(11.11)
Simple blood transfusion	
Yes	23(85.2)
No	4(14.8)
Exchange Transfusion	
Yes	6(25.9)
No	21(77.7)
ICU care(perioperative)	
Yes	4(14.8)
No	23(85.1)
Complications	4(14.8%)
Sickle cell related(pneumonia/?ACS)	1
Others	3
Death	0

*Includes one LSCS, one mitral valve replacement and one Hepaticojejunostomy

Table No.9 Classification

Variable	N (%)
Homozygous sickle cell disease	148(64.4)
Compound heterozygous sickle beta thalassemia	82(35.6)

Table No.10 Comparison Between homozygous sickle cell and sickle beta thalassemia

A. (Clinical parameters)

Varaible	**Sickle cell anemia(n-148) N(%)/Median(range)**	**Sickle beta thalasemia(n-82) N(%)/Median(range)**	**P value**
Demographic Profile			
Age (in years)			
at first symptom	4(1-54)	4(1-45)	0.5
at evaluation	14(1-54)	9(1-47)	0.03
Gender			
Male	91(61.5)	61(74.4)	
Female	57(38.5)	21(25.6)	
Clinical Profile			
Joint pain	123(83.1)	64(78)	0.37
Anemia	98(66.2)	54(65.8)	1.0

Avascular necrosis	23(15.5)	8(9.8)	0.33
Acute chest syndrome	12(8.1)	2(2.4)	0.14
cholilithiasis	12(8.1)	2(2.4)	0.14
Splenomegaly			
Yes	55(37.1)	60(73.1)	**0.00**
No	93(62.9)	22(26.9)	

Table No.11 Comparison Between homozygous sickle cell and sickle beta thalassemia

B. (Laboratory parameters at diagnosis)

Variable	Homozygous Sickle(n-148) Median(range)	Sickle beta thalassemia(n-82) Median(range)	p value
Hemoglobin	9.1(4.8-12.5)	8.6(4-11.9)	0.15
Mean corpuscular volume*	86.6(76-96.5)	70(57.7-79.2)	**0.000**
WBC count(x $x10^3$cells/mm^3)	11.5(2.1-39.6)	9.5(3.6-38.4)	0.003
Platelet count (x 10^3cells/mm^3)	267(11-783)	262(129-344)	0.7
Reticulocyte count	6.8(0.8-23.6)	5.2(0.6-23.7)	**0.001**
HbF(%)**	19.8(7-35)	19.2(8.9-54)	0.9
HbS(%)**	73.2(51-87)	70.4(45-78)	**0.001**

*MCV data was available in 142 of homozygous sickle cell patients and 80 sickle beta thalassemia patients after exclusion of post transfusion samples.

**HbF and HbS data was available in 120 patients of homozygous sickle cell anemia and 62 patients of sickle beta thalassemia after exclusion of post transfusion samples.

Table No.12 Comparison Between homozygous sickle cell and sickle beta thalassemia

(Use of hydroxyurea)

Variable	**Sickle homozygous (n-145) N(%)/Median(range)**	**Sickle beta thalassemia (n-77) N(%)/Median(range)**	**P value**
Hydroxyurea			
Yes	145(98%)	77(93.9%)	0.137
No	3(2%)	5(6.1%)	
Dose (mg/kg/day)	15(10-35)	15(10-30)	0.1
Adverse Effects	12(8.2%)	5(6.4)	0.6
Leucopenia	6	4	
Thrombocytopenia	3	1	
Aplasia	1	0	

Table No.13 Comparison Between homozygous sickle cell and sickle beta thalassemia

(Response to hydroxyurea)

Response At 6 months	n-142*		
CR	117(82.4)	53(68.8)	**0.04**
PR	23(16.2)	20(26)	
NR	2(1.4)	4(5.2)	
Response At last f/u			
CR	88(60.6)	43(55.8)	0.4
PR	55(37.9)	31(40.3)	
NR	2(1.4)	3(3.6)	

*****Data was not available for three patients at six month follow up

TABLE NO.14 FACTORS PREDICTING RESPONSE TO HYDROXYUREA

(Clinical Parameters)

Variable	**CR(N-131)** **N(%)/Mean ±SD**	**PR+NR(N-91)** **N(%)/Mean ±SD**	**P value**
Diagnosis			
Homozygous Sickle(n-145)	88(60.6)	57(39.4)	0.54
Sickle beta thalassemia(n-77)	43(57.1)	34(44.1)	
Age at presenting symptoms (years)	6.95(7.1)	6.21(7.1)	0.143
Gender			
Male(n-144)	87(60)	58(40)	0.112
Female(n-78)	45(57.7)	33(42.3)	
Symptoms			
Bone and joint pain	107(81.1)	76(83.5)	0.22
Symptomatic anemia	29(22.1)	16(17.5)	0.15
Pain and anemia	37(28.2)	26(28.6)	0.96
Avascular Necrosis	20(16)	11(12)	0.56

Table No 15 TABLE NO.14 FACTORS PREDICTING RESPONSE TO HYDROXYUREA

(Laboratory parameters- At diagnosis)

Variable	**CR(n-131)** **N(%)/ Median(range)**	**PR+NR(n-91)** **N(%)/ Median(range)**	**Pvalue**
Hemoglobin(g/dl)	9(4.5-11.9)	8.3(6.2-11.4)	**0.03**
Leucocyte ($x10^3$cells/mm^3)	10.2(2.1-396)	12.1(3.4-38.4)	0.06
Platelet ($x10^3$cells/mm^3)	233(129-653)	280(11-3449)	0.14
Reticulocyte (%)	5.8(0.68-23.6)	6.7(0.8-23.76)	**0.03**
Mean corpuscular volume (n-216)	82.5(57-108.2)	79.05(61.5-104.7)	0.21
Hb F(%) (n-182)	21(6.1-54)	17.6(4.7-33.3)	**0.01**
HbS(%) (n-182)	72.2(43-87)	72.8(38-87)	0.33
*HbF(%) (n-34)	32.05(20.5-51)	19.05(3.2-38.2)	**0.004**

*HbF at last follow up

Figure no 1. Approach to patient selection

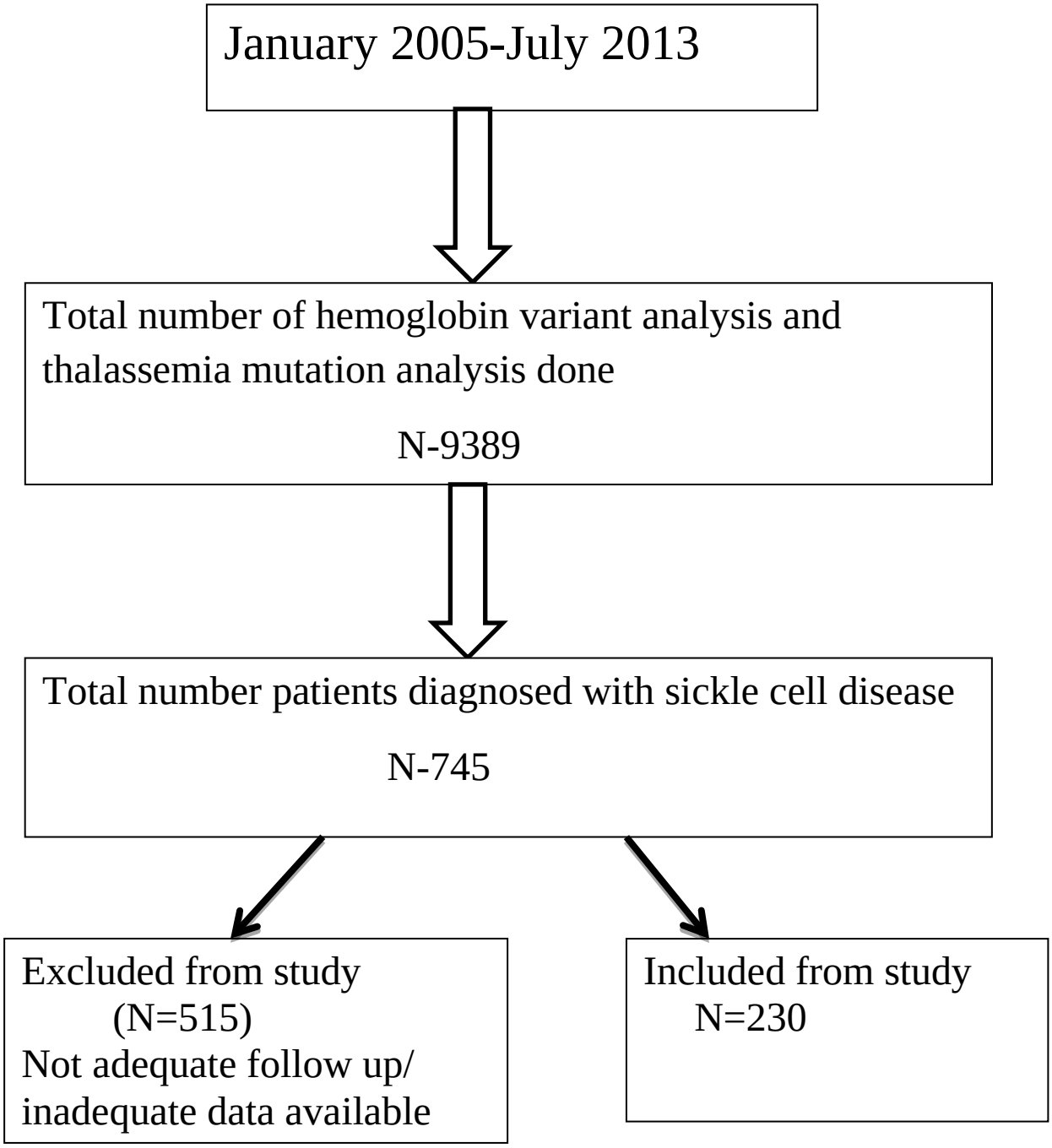

Figure No. 2. Distribution of patient population

Discussion

DISCUSSION

Sickle cell disease is a multisystem disorder with wide variations in presentation and severity ranging from being minimally symptomatic to transfusion dependent anemia or severe pain crises which can be fatal. In the present scenario, the role of hydroxyurea, and in certain situations, regular blood transfusions have been well established in improving the quality of life and overall survival. However in Indian patients, where the disease manifestations are usually mild and baseline HbF is higher, the ideal therapy is not well defined. Here an attempt was made to study the demographic profile and response to hydroxyurea treatment patients of sickle cell disease who were diagnosed and treated in Christian Medical College, Vellore from January 2005 - December 2013.

During the study period, 745 patients (7.9%) were diagnosed as sickle cell disease (including homozygous sickle cell anemia and compound heterozygous sickle beta thalassemia) out of 9389 patients evaluated for suspected hemoglobinopathy by hemoglobin variant analysis and thalassemia mutation analysis. This study includes 230 patients of sickle cell disease (out of 745 patients of sickle cell disease) who fulfilled the inclusion criteria of present study.

The median age of patients at evaluation was 12 years (range 1-54years), while median age of first symptom was 4 years (1-54years). The male: female ratio was 1.9:1 suggesting male preponderance. In comparison to other studies, our patients were older at the time of evaluation. In a study of 299 patients of sickle cell disease by Neonato et al (89), the median age at evaluation was 10.1 years (±5.8 years). In a study from a tertiary care hospital in south India by Chandra et al, the median age at diagnosis was 19.9 years(range3-48 years), while a study from

another Centre in Karnataka, India, the median age of diagnosis was 8.5 years(range 1-50 years).(90,91). The difference in age at evaluation could be due to referral bias as our study was carried out in tertiary care hospital with most of the cases belonging to other states. There were more male patients in our study which can be due gender bias prevalent in our country and has been noticed in other studies from India.

In our study, majority of the patients were from Eastern India with 72 patients (31.3%) from Jharkhand, 23 patients (10%) from Orissa and 23 patients (10%) from West Bengal. There were 36 patients (15.7%) from Tamil Nadu, 44 patients (19.1%) from Andhra Pradesh, 14 patients (6.1%) from Chhattisgarh and 2 patients (0.9%) from Bihar. In addition there were 14 patients (6%) from other states of India and two patients from Maldives. The various epidemiological studies have documented 0-18% prevalence of sickle gene in Eastern India. Although the central and western India is having high prevalence of sickle gene (22.5-44.4% and 0-33.5% respectively), the paucity of patients from these regions can be explained on the basis of referral bias. (92)

The bone and joint pain was the most common symptom in our study and was present in 187 patients (81.3%) and was the presenting symptom in 93 patients (40.4%). In 55 patients (24%), symptoms of anemia was presenting feature, out of these, 10 patients (4.3%) had transfusion dependent anemia requiring regular blood transfusions. In addition, 68 patients (29.6%) presented with both symptomatic anemia and joint pain at the time of diagnosis. Other rarer presentation at diagnosis included bone and joint pain with avascular necrosis in 11 patients (4.8%) and symptomatic anemia, pain along with avascular necrosis in two patients (0.9%). The overall frequency of symptomatic anemia was 54.3% (n-125) while anemia (as defined by WHO criteria) was present in 98.6% (n-227) of patients. The overall prevalence of other clinical

features included abdominal pain in 12.2 % (n-28), febrile episodes in 7.4% (n-17) and jaundice in 6.1% (n-14). Splenomegaly was present in 50% (n-115) of patients at diagnosis. In a study of 305 children of sickle cell disease diagnosed at birth, Bainbridge et al (93) noticed that acute painful episodes were most common presentation and was the most common symptom by two years of age and was present in 88% of patients. In a study by Chandra et al (90), 55 patients of sickle cell disease were prospectively studied and in their cohort of patients, symptoms at presentation included pain (80%), jaundice (85%) and anemia (60%).The splenomegaly was reported in 41% of their patients. The findings are similar to our study except for increased frequency of jaundice in their study. In another study from south India (Karnataka), Meera et al studied 60 patients of sickle cell disease and in their study anemia was most common symptom at presentation and was present in 83.3% of patients followed by bone pains present in 60% patients. (91) In comparison to our study, the symptomatic anemia was more prevalent than our cohort of patients while bone pains were less frequent as compared to our study. This can be explained both on the basis of phenotypic heterogeneity as sickle cell disease in known to have high phenotypic variability as well the patient population studied were different (East Indian versus natives of Karnataka).

Abdominal pain and fever was present in 28 (12.2%) and 17 (7.4%) patients respectively. In a study by Jain et al, 85 children with sickle cell disease were prospectively analyzed for morbidity pattern and acute febrile illness was noted in 30% of patients and was common morbid event during the study. (94).The higher incidence of fever in their study can be due to that only the children less than five years of age were included in the study that are more prone to infection. In addition, data on follow up may not have been fully captured during follow up.

Avascular necrosis was present in 13.9% of our patients. In literature, the incidence of avascular necrosis is known to be about 10 % (46). In study by Chandra et al, AVN was present in 5.4% of patients while in another study by Meera et al, only one patient out of sixty (1.6%) was found to have AVN (90,91). The higher incidence of AVN in our study population can be because of referral bias as many patients are referred for surgical intervention to our hospital.

Acute chest syndrome was present in 6.1% (n-14) of our patients. As compared to other studies from India, the incidence of acute chest syndrome has been reported in the range of 3.3% to 10.9%. (90, 94)

In our study, 167 patients (72.6%) had past history of blood transfusion at the time of diagnosis. This is higher as compared to study by Chandra et al (52% versus 72%), while in a study of pediatric patients with SCD by Jain et al, 85% of patients received blood transfusion.(90,94). The difference of blood transfusion practices is variable in our country and there are no specific guidelines for blood transfusion in sickle cell disease, which can account for the difference in frequency of blood transfusion in different studies.

In our study, cholilithiasis was documented in 6 patients (14%) as compared to 10% reported in a study by Tripathy et al in Orissa. (95). However, the difference could be due to that all patients in their study were screened for cholilithiasis with ultrasonography while in our study only symptomatic patients were screened.

Other rarer complications documented in our patients included leg ulcers in 1.3% (n-3), seizure disorder in 0.8% (n-2) and stroke in 0.8% (n-2). These complications are considered rare in Indian patients with sickle cell disease and a prevalence of <1% for stroke and leg ulcers has been reported in various studies. (94,21). In a study of Jamacian cohort of patients of sickle cell

disease, the cumulative incidence of seizures was reported as 2.2% , and in a study from Chhattisgarh India, seizure disorder was presenting symptom in 3.1% of patients.(36,96)

Majority of patients (125, 54.5%) in our study presented with symptomatic anemia and the median hemoglobin for entire cohort was 8.7g/dl (range: 4-12.5g/dl) at the time of diagnosis. 130 patients (56.5%) had leukocytosis (WBC count >10,000cells/mm^3) at presentation. These findings are similar to study by Chandra et al, with hemoglobin at diagnosis of 8.3g/dl and leukocytosis was present in 41.8% of their patients. (90) The baseline hemoglobin F was available for 182 patients and median HbF was 19.5 %(4.7-54%). This is higher as compared to data from western population (89) but is consistent with data from various Indian studies were HbF levels is known to be high. (94,97).

During this study period, 27 patients of sickle cell disease underwent different surgical procedures. The orthopedic surgeries were most common surgeries performed, while other surgeries included cholecystectomy, splenectomy , lower segment caesarean section, mitral valve replacement and hepaticojejenusotomy. 88.8% (n-24) of surgeries were managed with simple perioperative while in 25.9% (n-6) exchange transfusion was done. Most of the surgeries were uneventful and postoperative complications was reported in 4 patients (14.8%) and included pneumonia/acute chest syndrome in one patient while other patients had complications unrelated to sickle cell disease (hematemesis in one patient, increased blood loss during surgery in one patient and urinary tract infection in one patient) .Perioperative ICU care was required in 4 patients (14.8%) and there were no deaths reported. In a study by Sammak et al, retrospective analysis of 85 patients who underwent various surgical procedures were analyzed. In their study, 21.2% patients received exchange transfusion while 24.7% patients received simple blood transfusion in the preoperative period. In about half of the patients (54.1%) no blood transfusion

was given. The postoperative complications in study were 14.1% which is similar to our study. However 50% of their postoperative complications were due to vaso occlusive crisis and incidence of ACS varied between 5-11% between various types of surgeries performed (98). The decreased incidence of sickle cell related complications in our study could be due more liberal use of preoperative transfusion in sickle cell patients. In a multicenter study by Vichinsky et al (99), 118 patients of sickle cell disease who underwent 138 orthopedic surgical procedures were analyzed to assess perioperative complications and outcome. In their study, 67% of patients had serious perioperative complication, while 17% experienced sickle cell related complications most commonly acute painful crises or acute chest syndrome. There were two deaths in perioperative period. This is higher as compared to our study and may be due to smaller number of procedures in our study and different patient characteristics as sickle cell disease is considered to be more severe in western patients as compared to Indian patient.

Hydroxyurea was given in 222 patients (96.5%) at diagnosis while 8 patients (3.5%) where not started on hydroxyurea due to minimal symptoms at presentation. However, among these 8 patients, two patients were started on hydroxyurea later due to increased symptoms while six patients continue to be on follow up without the use of hydroxyurea. The initial dose of hydroxyurea was 10mg/kg/day in most patients and was increased in case of inadequate response by treating clinician. The median dose of hyroxyurea was 15mg/kg/day (range: 10-35mg/kg/day) with majority of the patients (171 patients, 77.1%) receiving ≤15mg /kg/day. The data from western studies have used doses of upto 35mg/kg/d and in most the studies the starting dose is 20-25mg/kg/day (83-86). However, in Indian patients, where baseline HbF is high and overall disease course is milder, the appropriate dose of hydroxyurea has not been well defined. In a study by Jain et al (100), low dose hydroxyurea (10mg/kg/day) was used in symptomatic

children with sickle cell disease and was found to be effective and safe. In a study by Patel et al (101) from Eastern India, low dose hydroxyurea (10mg/kg/day) was used in adults and children with sickle cell disease and was shown to be effective in both adult and pediatric patients with sickle cell disease.

The response to hydroxyurea therapy was assessed at six months and at last follow up. The median duration of follow up in our study was 36.25 months (range 6 -117months).

There was significant improvement in clinical and laboratory parameters after the treatment with hydroxyurea. The frequency of painful crisis per year decreased from 3.4 to 0.9 ($p<0.000$) while blood transfusion requirement also decreased significantly ($p<0.000$). Out of ten patients who presented with transfusion dependent anemia at diagnosis, four patients became transfusion independent after hydroxyurea therapy other five patients had marked decrease in their transfusion requirement. One patient continued to have regular blood transfusion requirement and it was probably because large splenomegaly in that patient. The occurrence of acute chest syndrome decreased significantly from 14 episodes (6.3%) at diagnosis to 4 episodes (1.8%) at last follow ($p=0.01$). The role of hydroxyurea in ameliorating the clinical manifestations of sickle cell disease have been well documented in various studies from West including the large Multi Centre Study of Hydroxyurea in Sickle cell anemia (MSH), a double blind randomized controlled trail, which established the efficacy of hydroxyurea treatment in sickle cell disease. In this study by Charache et al (80), 152 patients were treated with hydroxyurea while 147 patients were given placebo. The was significant difference in annual rates of painful crisis (2.5 versus 4.5), episodes of acute chest syndrome (25 versus 51) and decreased requirement for blood transfusion (48 versus 73) between the treatment group and control group.

In a study from India, Jain et al (100) treated 144 children with sickle cell disease with fixed dose of 10mg/kg/day of hydroxyurea for a median duration of 24 months. There was significant decrease in incidence of painful crises from 4.27 episodes/year to 0.15 episodes/year, acute chest syndrome (from 0.03 episodes/year to no episode during treatment), and blood transfusion requirement (0.77 units/year to 0.15 units/year). In another similar study by Singh et al (102), 24 adult patients of sickle cell disease were prospectively treated with hydroxyurea for one year with a median dose of 22mg/kg/day. At the end of one year, there was significant decrease in annual frequency of painful crisis (3.63 to 1.67) and hospitalizations (4.75 to 2.25) suggesting the efficacy of hydroxyurea in adult Indian patients with sickle cell disease. Our study findings are similar to above studies.

In our study, in addition to clinical improvement, the hematological parameters also showed significant improvement. The mean hemoglobin increased from 8.7±1.44 g/dl to 9.6±1.45 g/dl (p=0.000), while leucocyte count decreased from $11.71x10^3/mm^3$ (±5.5) to $9.0x10^3/mm^3$(p0.000). The HbF value at baseline and after treatment was available in 34 patients and showed in increase from 18.3% (±6.6) to 25.9(±10.06) (p=0.000). These observations are similar to other studies which have documented a significant increase in hemoglobin and HbF after treatment with hydroxyurea in Indian patients.(101,102).

Out of 230 patients, 222 patients who were treated with hydroxyurea urea and had at least six months follow up were included for response assessment. The response assessment was done after six months and at last follow up after initiation of hydroxyurea therapy.

The response was defined in terms of complete response, partial response and no response (as defined above). After a median follow up of 36.25 months, 131 patients (57.4%) had complete

response, 86 patients (37.4%) while 5 patients (2.2%) did not show any response to hydroxyurea therapy. In a study by Patel et al from Orissa, India (101), 118 patients were prospectively observed for response to hydroxyurea for duration of two years. In their study, 92.2% of adult patients and 71.5% of pediatric patients were termed as responders (defined as at least 50% reduction in painful crisis). This is high as compared to our study, but in our study complete response was defined as complete resolution of presenting symptom while partial response was defined as 50% reduction, so taken together(complete and partial responders;94.8%), the response rates are similar.

Hydroxyurea treatment was well tolerated in our cohort of patients with only 17 patients (7.6%) were documented to have adverse events. Most common side effects were hematological and include leucopenia in 10 patients (4.5%), thrombocytopenia in 4 patients and aplasia in one patient (0.9%). All events were mild and none required hospitalization or any other intervention except for dosage reduction of hydroxyurea. One patient who developed aplasia recovered counts after prolonged period was observation (about one year) and was asymptomatic on low dose of hydroxyurea (2.5mg/kg/day) at last follow up. These observations are similar to data from other Indian studies where mild hematological toxicity has been reported between 5-7%.(100,102) However, it is significantly less as compared hematological toxicity as reported in MSH study where almost all patients developed some degree of neutropenia and required temporary treatment cessation or decrease in dose. The difference can be due to the high dose used in this study where the target was to achieve maximum tolerated dose (MTD) of 35mg/kg/day. (80)

On categorizing patients into homozygous sickle cell disease and sickle beta thalassemia, 148 patients (64.4%) were having homozygous sickle cell disease were as 82 patients (35.6%) had

sickle beta thalassemia. There were no statistically significant differences in two groups with respect to median age first symptom (4 years in both) or gender distribution (male predominance in both groups). However median age at evaluation was lower in sickle beta thalassemia group as compared to homozygous sickle cell anemia (9 year versus 14years: p=0.03).There were no significant difference in frequency of painful episodes (83.1% versus 78%, p=0.37), anemia (66.2% versus 65.8%, p=1.0), avascular necrosis (15.5% versus 9.8%, p=0.33) or acute chest syndrome (8.1% versus 2.4%, p=0.1). However, splenomegaly was more commonly found in patients of sickle beta thalassemia as compared to homozygous sickle cell disease (73.1% versus 37.1, p=0.000). There is limited data comparing the clinical features of homozygous sickle cell anemia with compound heterozygous sickle beta thalassemia. However, in a study of 21 patients of sickle beta thalassemia by Mukherjee et al, similar clinical features were observed in their patient cohort as in our study.(103).

The comparison of laboratory parameters in these two groups of patient did not reveal significant difference with respect to mean hemoglobin, platelet count or HbF percentage at diagnosis. However, mean corpuscular volume, leucocyte count and reticulocyte percent were significantly low in sickle beta thalassemia as compared to homozygous sickle cell anemia. In addition median HbS percentage at diagnosis was higher in homozygous sickle as compared to sickle beta thalassemia. In our study, mean Hb for patients sickle beta thalassemia was 8.6 g/dl, mean corpuscular volume was 70fl , mean Hb F and HbS was 19.2% and 70.4% respectively. In a study of 21 patients of sickle beta thalassemia by Mukherjee et al (103), mean hemoglobin at diagnosis was 7.8 g/dl which is lower than observed in our study, while mean MCV was 73fl which is similar to our study. The higher hemoglobin in our patient population could be due to

blood transfusions given before reporting to our hospital. The HbS percent and HbF percent observed in their study were similar to our study.

On comparing the use and response of hydroxyurea in these two groups of sickle cell disease, there were no significant difference with respect to number of patients treated in each group (98% versus 93.9%, p=0.137), median dose of hydroxyurea used (15mg/kg/day in both group, p=1), or adverse events observed in these patients (8.2% versus 6.45,p=0.6). In addition, there was no significant difference in response to hydroxyurea treatment in these two groups as assessed at last follow up (complete respone:60.6% versus 55.8%, partial response:37.9% versus 40.3% p=0.4). The above observations suggest that hydroxyurea can be as effective and safe in sickle beta thalassemia as in homozygous sickle cell disease. In a study by Dehury et al (104) from Orissa, India, 203 patients of sickle beta thalassemia were prospectively treated with hydroxyurea (at a fixed dose of 10mg/kg/day) for a median duration of two years. They reported marked improvement with respect to annual frequency of vaso-occlusive crisis (3.5 to 0), blood transfusion (1 to 0) and hospitalization (1 to 0) in both adult and pediatric patients as observed in our study. However, they did not compare the observation with homozygous sickle cell disease patients.

A univariate logistic regression analysis of the common clinical and laboratory variables was done to detect predictors of complete response to treatment. This showed no significant association with age at presenting symptom (p=0.143), gender (p=0.112), homozygous sickle cell anemia versus sickle beta thalassemia (p=0.546), presenting symptom at diagnosis (p=0.154), leucocyte count (p=0.06), or platelet count (p=0.140) at diagnosis with the response status. However a significant association was observed with hemoglobin (p= 0.03), reticulocyte percent (p=0.03)and HbF percent (p=0.01) at diagnosis. It was observed that higher hemoglobin

and higher HbF percent at diagnosis are associated with higher rates of complete response at last follow up. A detailed literature search did not yield any studies that have analyzed clinical variables for detecting response to hydroxyurea, however, in a study by Jain et al (100), higher baseline HbF was associated with significantly higher increase in HbF after hydroxyurea therapy resulting in better clinical response. In our study, in patients who had complete response at last follow up were having higher HbF levels as compared to patients who had partial or no response (p=0.004). This observation is similar to the study by Patel et al (101), who observed that, after hydroxyurea therapy, HbF level was higher in responders as compared to non-responders.

LIMITATIONS OF STUDY:

1. Retrospective study: limited available data due to non-retrievable records.

2. This cohort of patients may not be truly representative of general population because of the referral bias, since most of patients presenting to the department were from few select states (catchment area of hospital) and not representative of general population.

3. Limited patient numbers and data available due to loss of follow up of patients

Conclusions

CONCLUSION:

1. Bone pains and anemia are the most common manifestations, while stroke priapism and leg ulcers are rare in Indian patients of sickle cell disease.
2. Homozygous sickle cell anemia and sickle beta thalassemia have similar clinical manifestations.
3. Low dose hydroxyurea (up to 15mg/kg/day) is safe and efficacious in ameliorating the symptoms in both the patients of homozygous sickle cell anemia and sickle beta thalassemia.
4. Surgical procedures can safely be performed in patients of sickle cell disease.
5. Some patients of sickle cell disease, albeit very few, may not require treatment with hydroxyurea and can be followed with regular monitoring.
6. Large prospective trials are required to form the guidelines for use of hydroxyurea in Indian patients with sickle cell disease addressing the issues of indications, dosage and monitoring.

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Appendix

name	hosp no	age at dx	age at c	gender	Diagnosis-cmc	Dx	date of Dx	family history	presenting :	Age at pres	Age at fi	any oth	any surgery d	Any e	compl	Treatment dose	duration	complications	any other co	Hb at I
		at cmc	outside	1-male	1-Homozygous SC	outside	.	1-yes	1- joint pain	.	.	abd pair	1- yes	1yes	.	1- hydroxy	.	1-leucopenia	.	.
		.	.	2-female	2-Compound B-sc	.	.	2-no	2- Transfusi	.	.	3-cholili	2-no	2-no	.	2-nil	.	2- thrombocyt	.	.
		.	.	.	.	.	.	.	.	.	.	5-jaundi	.	3-tran	.	.	.	3-aplasia,4-Hy	.	.
CHUMKI PRADHAN	188250D	21	na	2	1	na	14/02/2008	2	8	16	20	nil	1	3	nil	1	15	nil	nil	9.5
SHAMSHAD BEGUM	211678D	45	na	2	2	na	01/04/2008	2	7	45	45	nil	2	na	na	1	20	2	1 nil	5.9
Golam Rabbani	040038D	6	4	1	1	1	06/06/2007	2	3	1	na	nil	2	na	na	1	15	7	nil	constipation	8.5
Nasir shaik	069442D	15	14	1	1	2	24/07/2007	2	7	14	14	1	2	na	na	1	20	5	nil	nil	6
Mathiyarasan	251164D	13	na	1	1	na	06/06/2008	2	7	3	12	1	2	na	na	1	10	5	nil	nil	7.6
Ramesh K	283477D	9	na	1	1	na	29/07/2008	2	1	1	9	6	2	na	na	1	15	5	nil	nil	5.4
Ayisa S	537891C	8	na	2	2	na	15/07/2005	2	1	5	5	nil	2	na	na	1	20	9	nil	nil	7.9
Sitaram Panika	305956D	9	7	1	1	1	10/09/2008	1	7	7	7	1	2	na	na	1	15	5	nil	nil	9.4
Mohammad Kaif	729074C	8	2	1	1	1	10/11/2005	2	1	1	na	nil	2	na	na	1	20	8	nil	nil	7.7
Himanshu Manji	873740C	4	2	1	1	1	11/08/2006	2	1	2	na	nil	2	na	na	1	15	7	nil	nil	8
Akash Kumar	356854D	9	9	1	1	1	21/11/2008	2	7	3	3	nil	2	na	na	1	15		nil	nil	8.5
Arti Singh	359287D	35	35	2	1	1	25/11/2008	2	1	25	na	nil	2	na	na	1	20		nil	nil	9
Ramisetti ram narayan	653372C	7	7	1	1	1	14/06/2005	1	1	6	na	nil	2	na	na	1	20		nil	nil	9.1
Prakash Kumar	189716D	16	3	1	1	1	18/02/2008	2	7	3	3	4	1	1	nil	1	15		nil	nil	6.2
Hezekiya	191297D	4	3	1	2	1	27/02/2008	2	7	3	3	nil	2	na	na	1	20		nil	nil	7.1
Arthy	013724D	4	na	2	1	na	24/12/2008	2	4	1	1	nil	2	na	na	1	25	4	nil	nil	7.5
Mahtab Alam	446660D	4	4	1	1	1	20/04/2009	2	1	2	na	nil	2	na	na	1	20		nil	nil	9.3
Rishu Yadav	453641D	5	3	1	1	1	01/05/2009	2	4	3	5	nil	2	na	na	1	20		nil	nil	8.3
Anil Mohan	470250D	25	15	1	1	1	29/05/2009	2	1	3	3	1	1	3	nil	1	10		nil	nil	9.1
hema prabha	470986D	12	11	2	2	2	12/06/2008	2	7	11	11	nil	2	na	na	1	10		nil	nil	7.3
Susoritha	517921D	34	30	2	1	1	21/08/2009	2	7	5	6	6	2	na	na	1	20		nil	nil	8.8
santosh kumar	584281D	19	10	1	1	1	14/11/2009	2	7	3	6	4	1	3	nil	1	10		nil	nil	8.8
Gondela swathi	595387D	16	12	2	1	1	10/02/2010	2	1	12	15	3	2	na	na	1	15		nil	nil	9
Deepti Rekha	594265D	54	na	2	1	na	15/12/2009	1	4	54	na	3	2	na	na	1	10		nil	nil	7.3
Shamim MD	526962D	8	na	1	2	na	22/08/2009	2	7	8	na	1	2	na	na	1	15		nil	nil	8.1
Naveen P	562828c	9	5	1	2	1	07/01/2005	2	7	1	na	2	2	na	na	1	10		nil	nil	8.3
Mansi	401986D	4	2	2	2	2	17/02/2009	2	4	2	2	nil	2	na	na	1	15		nil	nil	10.5
Suman Maity	632161D	11	5	1	2	1	10/02/2010	2	7	5	5	nil	2	na	na	1	15		nil	nil	7.1
Meyyalagan	637199D	7	6	1	1	1	12/02/2010	2	7	5	5	nil	2	na	na	1	15		nil	nil	8
Siva kumar	543180D	17	10	1	1	1	21/09/2009	2	1	10	15	nil	2	na	na	1	10		1	nil	9.5
Akanksha Sahu	641695D	21	2	2	1	1	21/02/2010	2	7	2	2	3	1	3	nil	1	10		nil	nil	6.7
Mukilan B	672568C	1	na	1	2	na	22/07/2005	1	10	1	1	4	2	na	na	1	10		nil	nil	7.1
Santanu Roy	720432D	5	4	1	2	1	06/08/2010	2	4	4	4	nil	2	na	na	1	10		nil	nil	9.6
Jasmin Rexna khora	751448D	28	5	2	1	1	18/08/2010	2	10	5	14	3	1	3	na	1	15		nil	nil	9.5
Atanu Roy	756071D	1	na	1	2	na	09/08/2010	2	4	1	1	nil	2	na	na	1	15		nil	nil	7.9
philip Mathew	772923D	13	12	1	1	1	02/09/2010	2	7	2	2	7	1	3	na	1	10		nil	nil	7.5
Murali M	771155D	16	na	1	2	na	01/09/2010	2	1	12	na	1	2	na	na	1	10		nil	nil	8.9
Sesi bhushan k	766480D	21	12	1	2	1	23/08/2010	2	1	3	3	1	2	na	na	1	10		nil	nil	10.2
Vikash Kurrey	812563D	28	5	1	1	1	28/10/2010	1	11	5	5	nil	2	na	na	1	10		nil	nil	8.6
Sarathi K	798979D	7	na	1	1	na	04/12/2010	2	4	1	1	4	2	na	na	1	10		nil	nil	6.9
Balaji Naik	749257D	22	18	1	2	1	24/08/2010	2	7	15	15	nil	1	3	nil	1	20		nil	nil	11.3
gunasrija	867176D	1	1	2	1	1	11/01/2011	2	1	1	na	nil	2	na	na	1	10		nil	nil	9.8
Sheikh Sultan	843311D	24	na	1	1	na	04/01/2011	2	1	3	na	nil	2	na	na	1	10		nil	nil	10.3
Sohel hussain	925894D	20	na	1	1	na	10/05/2011	2	4	4	4	3	2	na	na	1	10		nil	nil	6.9
mohammad azad K	935488D	7	6	1	2	1	21/05/2011	2	1	6	na	nil	2	na	na	1	10		nil	nil	7.1
Prafulla Sahu	976624D	24	3	1	1	1	29/06/2011	2	8	3	22	nil	1	3	nil	1	20		nil	nil	9.4
Hari Prasanth	003566f	6	5	1	2	1	29/08/2011	2	4	5	5	nil	2	na	na	1	15		nil	nil	8.4
Pawan Kumar	000503F	5	3	1	2	1	03/08/2011	1	7	3	3	nil	2	na	na	1	10		nil	nil	9.8
rahamatullah	985608D	29	17	1	2	1	06/09/2011	1	1	17	na	nil	2	na	na	1	20		nil	nil	8.9
Bharat J	038970F	3	3	1	2	1	20/08/2011	2	7	3	na	nil	2	na	na	1	15		nil	nil	6.8
Saroj Devi	037746F	24	10	2	1	1	21/09/2011	2	8	10	na	6	1	3	nil	1	10		nil	nil	9.6
Satish Kumar	017115F	51	35	1	1	1	05/10/2011	2	1	5	50	nil	2	na	na	1	10		nil	nil	10.2
Ramya C	957830D	24	7	2	1	1	03/06/2011	2	1	6	6	4	2	na	na	1	10		nil	nil	8.1
Basanti gadi Shyam	084348F	25	15	2	2	1	09/12/2011	1	1	15	20	nil	2	na	na	1	10		nil	nil	10
Mirnalini	101191F	1	1	2	1	1	20/12/2011	2	4	1	na	nil	2	na	na	1	10		nil	nil	8.1
Baidnath Malakar	894599D	14	12	1	1	1	01/03/2011	2	1	5	10	nil	2	na	na	1	20		nil	nil	8

snigdha Das	840709D	12	3	2	1	1	07/12/2011	1	1	3	3	nil		2	na	na	1	15		nil	nil		9.5
Ram Bharat Singh	476079D	32	22	1	2	1	16/06/2009	2	9	18	18		3	2	na	na	1	15		nil	nil		8.3
Esther Hannan G	003393F	13	5	2	2	1	19/08/2011	1	7	5	7	nil		2	na	na	1	12		nil	nil		9.6
degala Prasad	042086F	9	5	1	1	2	27/09/2011	2	4	5	5		1	2	na	na	1	15		nil	nil		8.3
Mohammad Uzair	011062F	4	4	1	2	1	28/10/2011	2	7	4	4		1	2	na	na	1	15		nil	nil		8.2
Nishant Chandra	076180F	6	4	1	1	1	15/11/2011	2	7	4	4	nil		2	na	na	1	15		nil	nil		10.3
Nikhil Khora	084343f	6	5	1	1	1	25/11/2011	2	7	4	4		4	2	na	na	1	20		nil	nil		7.3
Mamidi Shanmukha	083243F	16	15	1	1	1	09/12/2011	2	4	4	7		6	2	na	na	1	15		nil	nil		7.9
Priya Nair	051989F	22	12	2	1	1	12/01/2012	2	11	12	12	nil		2	na	na	1	15		nil	nil		9.9
Samarat kumar	105314F	4	1	1	1	1	13/01/2012	2	4	1	3	nil		2	na	na	1	30		nil	nil		6.2
Sankar Naik	132496F	22	12	1	2	1	27/02/2012	2	1	12	12	nil		2	na	na	1	10		nil	nil		9.8
Baibhab Patra	146404F	1	1	1	1	1	25/02/2012	2	1	1	na	nil		2	na	na	1	35		nil	nil		7.4
Nisha Kumari	170577F	9	7	2	1	1	10/04/2012	1	7	3	3	nil		2	na	na	1	15		nil	nil		8.2
Jawaharlal M	180358F	24	20	1	1	1	19/04/2012	2	1	15	15		1	2	na	na	1	15		nil	nil		8.9
Pankaj Singh	181529F	28	3	1	2	1	22/05/2012	2	7	3	5		7	2	na	na	1	15		nil	nil		8.3
Anitesh Shit	270250F	11	1	1	2	1	14/08/2012	2	1	3	na	nil		2	na	na	1	20			2	nil	9.6
Chaitanya Naik	272840F	6	4	1	2	1	28/08/2012	2	7	2	2	nil		2	na	na	1	15		nil	nil		7.8
Rashmi Rose	272381F	37	30	2	2	1	04/09/2012	2	7	30	30		6	2	na	na	1	10		nil	nil		8
Uthra Kumari	828413A	26	20	2	1	1	17/07/2012	1	1	12	12	nil		2	na	na	1	15		nil	nil		7.1
Sk Niyamat	317565F	18	na	1	1	na	23/11/2012	2	7	6	6	nil		2	na	na	1	15		nil	nil		9.3
Kundan Mukhi	290724F	35	20	1	1	1	09/10/2012	2	1	12	12		3	1	1	nil	1	10		nil	nil		8
Megha Mahpatra	319938F	6	na	2	2	na	11/10/2012	2	1	6	6		1	2	na	na	1	15		nil	nil		7
Venkat B	325328F	5	4	1	1	1	02/11/2012	2	7	4	5	nil		2	na	na	2	na		na	na		8.8
Surya Kanth	330502F	5	2	1	1	1	16/11/2012	2	7	2	2	nil		2	na	na	1	15		nil	nil		9.7
Supriya G	324118F	12	3	2	1	1	25/10/2012	2	4	1	1	nil		2	na	na	1	15		nil	nil		8.4
Lingam Sonu	343284F	16	16	1	1	1	18/12/2012	2	1	16	na	nil		2	na	na	2	na		nil	nil		11.4
Juhi Urshala	141377F	5	4	2	1	1	02/03/2012	2	1	3	3	nil		2	na	na	1	10		nil	nil		11.3
Sanjay	184231F	2	2	1	2	1	18/04/2012	2	4	2	2	nil		2	na	na	1	15		nil	nil		8.5
Abhishek P	195777F	6	na	1	1	na	16/05/2012	2	7	3	3	nil		2	na	na	1	15		nil	nil		10.9
Palak Kumari	202784F	9	7	2	2	1	01/06/2012	2	7	5	5	nil		2	na	na	1	20		nil	nil		8
Rounak Kumar	217489F	3	3	1	1	1	03/07/2012	2	4	3	3	nil		2	na	na	1	15		nil	nil		9.4
Mohammad Nadim	284076F	4	4	1	2	2	14/09/2012	2	4	4	4	nil		2	na	na	1	15		nil	nil		8
Vijayasree	283390F	4	4	2	1	1	14/09/2012	2	1	4	4		1	2	na	na	1	10		nil	nil		10.3
jogeshwar bhakat	233760F	14	5	1	2	1	26/06/2012	2	4	5	4		1	2	na	na	1	10			1	nil	7.4
Arbaz Khan	302193F	10	7	1	2	2	05/10/2012	2	7	7	7		1	1	3	na	1	20		nil	nil		6.3
Ram roshan	320344F	4	3	1	1	1	26/10/2012	2	7	3	3	nil		2	na	na	1	10		nil	nil		9.4
Pratibha	328055F	9	7	1	2	1	26/10/2012	2	7	6	6		1	2	na	na	1	10		nil	nil		8.2
Badre Alam	879287D	28	na	1	1	na	05/02/2011	2	3	5	12		6	1	1	nil	1	20		nil	nil		11.3
Swarna Mahnt	398956F	13	11	2	1	1	01/02/2013	2	4	7	7		6	2	na	na	1	20		nil	nil		8.9
Subash Karmakar	936077D	21	12	1	1	1	03/05/2011	2	1	10	na	nil		2	na	na	1	25		nil	nil		9.6
Tarani	400201F	7	5	2	1	1	15/02/2013	2	7	2	2	nil		2	na	na	1	10		nil	nil		8
Rahul raj Baraik	407825F	12	4	1	1	1	14/02/2013	2	4	4	4		3	2	na	na	1	10		nil	nail hyperpig		8
Ayush Kumar	414352F	12	3	1	2	2	01/03/2013	2	7	3	10	nil		1	1	nil	2	na		nil	nil		6.2
Joy Nayek	391249F	10	2	1	1	1	26/02/2013	2	1	2	na	nil		2	na	na	1	10		nil	nil		8.5
Gopika	420804F	4	3	2	2	1	09/04/2013	2	1	3	na	nil		2	na	na	1	15		nil	nil		10.7
Varsha Chauhan	427663F	5	4	2	2	1	19/03/2013	2	7	3	3		1	2	na	na	1	20		nil	nil		7.3
Vasu pali	426624F	12	7	1	2	1	26/03/2013	2	7	7	7	nil		2	na	na	1	10		nil	nil		11.2
Srilatha	411243F	13	4	2	2	1	02/04/2013	1	7	4	4	nil		2	na	na	1	10		nil	nil		9.9
Dewashish kumar	491184F	11	3	1	1	1	28/05/2013	2	11	3	3	nil		2	na	na	1	15			1	nil	8.6
Adarsh Kumar	449837F	3	3	1	2	1	03/05/2013	2	4	3	2	nil		2	na	na	2	na		na	na		4
Sarwan Kumar	608561F	34	30	1	1	1	25/06/2013	2	1	24	24	nil		2	na	na	1	25		nil	nil		6.8
Rathina Vel	496877f	14	na	1	1	na	10/06/2013	2	4	12	12	nil		2	na	na	1	15		nil	nil		6
Akhila S	616502F	10	6	2	1	1	19/07/2013	2	1	6	na	nil		2	na	na	1	15		nil	nil		8.6
Mahesh Nimala	636246F	18	12	1	1	1	16/08/2013	2	1	10	na	nil		2	na	na	1	15		nil	nil		11.4
Ravichandran	617678F	20	na	1	2	na	04/07/2013	2	1	16	na		1	2	na	na	1	10		nil	nil		8.4
Kamal Hussain	650657C	11	7	1	1	1	07/06/2005	2	7	2	1	nil		2	na	na	1	10		nil	nil		10.6
Sidhardha	635661F	5	5	1	2	1	23/08/2013	2	4	3	3	nil		2	na	na	1	20		nil	nil		9.1
Aman Kumar	660512F	7	4	1	1	1	27/08/2013	2	4	4	na		5	2	na	na	1	20		nil	nil		8
Roushani Kumari	660300F	9	9	2	2	1	10/09/2013	2	1	3	na		1	2	na	na	1	10		nil	nil		9.8
Debasis Gouda	652288F	7	4	1	1	2	13/09/2013	1	1	4	na		1	2	na	na	1	15		nil	nil		9.6

Nilandri Gouda	652286F	10	6	2	1	2	13/09/2013	1	1	6	7	nil		2	na	na	1	15		nil	nil	10.2
Murali N	145581F	22	na	1	1	na	29/03/2012	2	1	1	10	nil		2	na	na	1	25		nil	nil	9.4
Rabiul Mondal	683802F	28	na	1	1	na	24/09/2013	2	7	18	24		5	2	na	na	1	10		nil	nil	8.3
Smriti verma	660788F	32	6	2	1	1	14/09/2013	2	8	6	12		4	1	3	nil	1	15		nil	nil	9.1
Kanak Prakash	638400F	14	6	1	1	1	08/10/2013	2	7	6	6		5	2	na	na	1	15		nil	nil	8.4
Rohith K	685214F	5	2	1	1	1	11/10/2013	2	1	2	3	nil		2	na	na	1	10		nil	nil	9.2
Koruvada Jyoti	722522f	26	26	2	1	1	26/11/2013	2	1	20	22	nil		2	na	na	1	20		nil	nil	7.8
Nigar Tirkey	729710F	36	36	2	1	1	10/12/2013	2	4	30	na		5	2	na	na	1	10		nil	nil	9.5
Bushra Arsh	768016F	4	2	2	1	1	20/12/2013	1	1	2	na	nil		2	na	na	1	15		nil	nil	9.7
Hammad Nasim	768015F	9	6	1	1	1	20/12/2013	1	1	1	na	nil		2	na	na	1	15		nil	nil	8.6
Avimanu Kumar	445268F	14	10	1	1	1	09/04/2013	2	1	3	7	nil		2	na	na	1	15			1	nil	10.8
Brinda	479062F	5	3	2	2	1	04/06/2013	2	4	2	2	nil		2	na	na	1	15		nil	nil	7.9
Abhey Pandey	622046F	13	13	1	2	2	26/07/2013	2	1	13	na	nil		2	na	na	1	10		nil	nil	9.8
Kalam Ansari	670766F	7	6	1	2	2	01/10/2013	2	7	3	3	nil		2	na	na	1	15		nil	nil	9.8
Sambasiva Rao	656969F	24	14	1	1	1	10/09/2013	2	8	14	14	nil		1	3	nil	1	15			2	nil	10.9
Arpit Singh	693867F	7	7	1	1	1	29/10/2013	2	7	4	4	nil		2	na	na	1	10		nil	nil	8.3
deepshikha Tirkey	711168C	12	12	2	1	1	10/10/2005	2	7	5	5	nil		2	na	na	1	25		nil	nil	6.4
Sourav Kumar	810004C	3	3	1	1	1	20/04/2006	2	1	2	3		1	2	na	na	1	20		nil	nil	10.2
indrajeet patra	919991c	24	12	1	1	1	30/10/2006	2	7	12	12		1	2	na	na	1	20		nil	nil	9.6
tarun Shankar	980186C	19	na	1	1	na	16/02/2007	1	7	1	3	nil		2	na	na	1	20		nil	nil	10.6
lingam sujata	816973C	19	10	2	1	1	05/05/2006	2	7	10	10		1	1	3	nil	1	20		nil	nil	7
Ranjeeth	750810C	7	na	1	1	na	27/12/2005	2	1	2	5		9	2	na	na	1	15		nil	nil	9.4
Chandini D	065934D	8	8	2	1	1	17/07/2011	1	7	4	5		2	2	na	na	1	15		nil	nil	6.8
Santosh Pradhan	188241D	19	12	1	1	1	19/02/2008	1	1	12	12	nil		2	na	na	1	10		nil	nil	9.5
Mamta Yadav	179256D	29	20	2	1	1	22/02/2008	2	1	20	20		3	1	2	na	1	10			2	nil	10.2
Nadeem	193204D	3	na	1	1	na	26/02/2008	2	7	3	3		2	2	na	na	1	15		nil	nil	10.9
Devashish Goswami	291404B	47	34	1	2	1	20/05/2008	2	1	15	na		3	2	na	na	2	na		na	na	10.2
Upasna Poorna	898415B	26	5	2	1	1	20/07/2008	2	7	5	12	nil		2	na	na	1	15			4	nil	10.4
Moureshwara	096525D	4	4	1	1	1	01/09/2007	2	1	3	na	nil		2	na	na	1	20		nil	nil	7.8
Rishana	248992D	7	3	2	1	2	30/05/2008	2	4	3	3		2	2	na	na	1	15		nil	nil	10.7
sujan baraik	333754D	19	na	1	1	na	13/10/2008	2	4	15	5		3	1	2	na	1	10		nil	nil	9.9
Mohammad Rafik	364243D	28	22	2	1	1	05/11/2008	2	1	3	15		3	1	3	nil	1	10		nil	nil	8.6
Kanishk	201280D	4	4	1	2	1	11/04/2008	2	4	3	3	nil		2	na	na	1	15		nil	nil	8.5
priyanshu	201117D	8	1	2	1	1	12/03/2008	2	9	1	1		1	2	na	na	1	15		nil	nil	8.1
Simran Taj	274432D	7	7	2	1	2	18/07/2008	2	4	1	2		2	2	na	na	1	15		nil	nil	9.1
Mohammad Furqan	277711d	5	4	2	2	2	21/07/2008	2	4	4	4		1	2	na	na	1	15		nil	nil	8.2
Akash Gorai	280801D	9	na	1	2	na	25/07/2008	2	4	7	8		2	2	na	na	2	na		na	na	9
Sandeep	322290D	4	2	1	2	2	24/09/2008	2	4	2	2		2	2	na	na	1	15		nil	nil	7.1
manik ghashi	321588d	26	23	1	1	1	30/09/2008	2	1	3	25	nil		2	na	na	1	10		nil	nil	11.4
sahil sahu	381947d	9	3	1	2	2	09/01/2009	2	7	2	3		1	2	na	na	1	15		nil	nil	8.9
surya durga prasad	384636D	19	3	1	2	2	05/01/2009	2	1	3	18		5	2	na	na	1	10		nil	nil	11.9
lokesh kumar	304843d	23	18	1	1	1	30/08/2008	2	8	12	22		4	1	1	na	1	10		nil	nil	11.5
draupadi Patra	461111D	23	7	2	1	1	15/05/2009	2	4	7	7	nil		2	na	na	1	15		nil	nil	5.5
Swagatika bhuyan	477326d	14	nd	2	1	nd	09/06/2009	2	8	10	10	nil		2	na	na	1	10		nil	nil	9.5
kamal uddin ansari	489966d	22	21	1	1	1	29/06/2009	2	8	3	21		10	2	na	na	1	10		nil	nil	7.4
kala rani	519686d	27	24	2	1	1	18/08/2009	2	1	3	7		5	1	3	na	1	20		nil	nil	9.6
neha khatoon	506030d	10	9	2	1	1	31/07/2009	2	4	9	6	nil		2	na	na	1	15		nil	nil	7.6
Srikanth	396787D	9	5	1	2	1	06/02/2009	2	9	5	4		5	1	3	nil	1	15		nil	nil	9.8
kumar Ayush	652379C	2	na	1	2	na	11/06/2005	2	4	1	na		5	2	na	na	2	na		nil	nil	9.4
Kinche giridhar	425708d	8	7	1	1	1	24/03/2009	1	4	7	7		2	2	na	na	1	15		nil	nil	6.5
janardan kumar	633273d	18	9	1	1	1	06/04/2010	2	1	9	15		2	2	na	na	1	20		nil	nil	9.9
samaresh maity	636432d	8	3	1	2	2	16/02/2010	1	1	3	na	nil		2	na	na	1	12		nil	nil	7.8
priyanka kumari	626818c	24	5	2	1	1	19/04/2005	2	1	3	15		4	1	3	nil	1	15			1	nil	10.7
lavanya N	641794D	14	nd	2	1	nd	16/04/2010	2	1	3	5			2	na	na	1	15		nil	nil	7.2
Anup kumar sahu	645331d	13	na	1	2	na	26/02/2010	2	1	3	na		6	2	na	na	1	15		nil	nil	9.7
kavitha S	681073d	31	nd	2	1	nd	20/04/2010	1	1	nd		25	nil	1	3	nil	1	20		nil	nil	9.9
Archana kumari	636836d	17	15	2	1	1	09/03/2010	2	1	15	na	nil		2	na	na	1	15		nil	nil	10.6
Dhash Chourey	613785D	4	1	1	2	1	03/08/2010	1	7	1	1	nil		2	na	na	1	15		nil	nil	7.7
Rathi D	752782D	22	6	2	1	1	10/08/2010	1	1	6	15		2	2	na	na	1	15		nil	nil	9.3
Sridhar	807073D	14	9	1	1	1	15/10/2010	2	1	9	na	nil		2	na	na	1	15		nil	nil	9.2

Ahmed Aslam	811759D		29	21	1		2	2	22/10/2010	1	1		3	na		nil			2	na		na	1	15			nil		nil		9
Bharati kumari	814371D		17	14	2		1	1	26/11/2010	2	1		14		10		8		1		3	na	1	15			nil		nil		9.4
jahangir Alam	848257D		24	13	1		1	1	11/01/2011	1	1		13		13	nil			2	na		na	1	10			nil		nil		11.6
Gadi Ram sai prasad	700391D		9	1	1		2	2	20/05/2010	2	7		1		1	nil			2	na		na	1	30			nil		nil		7.7
Tapas Mohanty	792234D		28	15	1		2	2	23/09/2010	2	7		7		14	nil			2	na		na	1	10			nil		nil		8.9
Mangel Patel	848222D		20	10	1		2	1	11/01/2011	2	7		3		15	nil			2	na		na	1	15			nil		nil		9.1
maryam moosa	854556d		22	19	2		1	1	14/01/2011	1	1		19	na		nil			2	na		na	1	15				4	nil		9.5
Krishnaveni	863070d		37	nd		2	1	nd	13/01/2011	1	1		3	na			3		1		3	wounc	2	na			na		na		10.1
Raj kumar M	854913D		16	na		1	2	na	01/02/2011	2	1		13	na			10		2	na		na	1	10				1	nil		9.7
Pratikshya patel	906604d		11		5	2	1		1	29/03/2011	2	8		6	na		nil		1		3	nil	1	15			nil		nil		9.6
Anas Ansari	821540D		1	na		1	2	na	03/04/2011	2	4		1		1		2		2	na		na	1	20			nil		nil		7.9
Rohan tanty	893441D		17	na		1	2	na	09/04/2011	2	1		3		10		5		2	na		na	1	15			nil		nil		11
Lakshmi kumari	944730D		11		9	2	1		1	18/05/2011	2	4		7		7		1	2	na		na	1	15			nil		nil		9.8
urvashi kumari	960662D		20		19	2	1		1	01/07/2011	2	4		3		19		3	1		3	nil	1	12.5			nil		nil		4.8
Mitali Singh	988760C		3	na		2	1	na	24/08/2011	2	1		2		3		1		2	na		na	1	15			nil		nil		7.3
Samiruddin ansari	034065F		30		20	1	2		1	20/09/2011	2	1		20		20		5	2	na		na	1	10			nil		nil		9.3
Prince kumar	092654f		18		6	1	2		1	16/12/2011	2	1		5		5	nil		2	na		na	1	15			nil		nil		9.5
Jithenra	940028D		1		1	1	1		1	10/05/2011	2	4		1		1		2	2	na		na	1	15			nil		nil		6.9
mitlesh kumar	086795D		29	nd		1	1	nd		21/08/2007	2	1		3		20	nil		2	na		na	1	15				1	nil		10.3
Subrata das	964237D		28	na		1	2	na		06/09/2011	2	4		15		27		1	2	na		na	1	10			nil		nil		7.1
nidhi mandavi	052038F		16	nd		2	2	nd		15/02/2012	2	9		7		7		8	2	na		na	1	20			nil		nil		5.8
Aryan lima	084121F		8		3	1	1		1	25/11/2011	2	7		1		2	nil		2	na		na	1	15				1	nil		8.2
Bollam viswahith	064379F		2		2	1	1		1	06/12/2011	2	4		1		1	nil		2	na		na	1	15			nil		nil		11.9
Rabindra Shasni	157605F		20		9	1	1		1	12/03/2012	2	7		9		20	nil		2	na		na	1	10			nil		nil		7.8
rajasri M	263420F		4		3	2	2		2	31/08/2012	2	1		3	na		nil		2	na		na	1	12			nil		nil		8.6
suraj koiri	292289F		13	na		1	2	na		06/09/2012	2	6		2		13		6	2	na		na	1	15			nil		nil		6.3
Nisha Kumari	307193F		15		11	2	2		2	12/10/2012	2	7		4		8	nil		2	na		na	1	25				1	nil		8.6
bikash patel	186068F		21		1	1	1		1	27/04/2012	1	1		3		6	nil		2	na		na	1	15			nil		nil		8.2
rhadeshayam	342195F		17		6	1	1		1	20/11/2012	1	7		6		6		9	2	na		na	1	20			nil		nil		9.8
Shakeel ahmed	353456F		24		16	1	1		1	24/11/2012	1	1		16		16	nil		1		1	nil	1	15			nil		nil		10.8
dilip kumar sahu	123321F		47		3	1	1		1	24/01/2012	2	1		2		25		6	1	na		na	1	15				3	nil		6.2
Alka Shewta	217161F		29		20	2	2		1	08/06/2012	1	7		19		20	nil		2	na		na	1	10			nil		nil		8.9
Sovika Das	164092F		12	na		1	2	na		08/06/2012	2	1		12	na		nil		1		3	nil	1	10			nil		nil		10
ritika Kumari	248905F		8		6	2	2		2	19/07/2012	2	7		5		5	nil		2	na		na	1	15			nil		nil		9.9
jaharul Sarkar	285586F		24		14	1	1		2	04/09/2012	2	1		14		14		2	2	na		na	1	20				2	nil		7.8
javed Akhter	361222F		17	na		1	2	na		18/12/2012	2	4		16		16	nil		2	na		na	1	10			nil		nil		9.7
Siddhardha	293939F		6		3	1	1		2	16/10/2012	2	7		3		3		2	2	na		na	1	20			nil		nil		10.9
golluri nandini	293932F		15		4	2	1		2	16/10/2012	2	4		4		4	nil		2	na		na	1	20			nil		nil		10.4
Anshul L	383511F		9		2	1	2		1	08/01/2013	2	1		2		2		2	2	na		na	1	20			nil		nil		8.8
Hemanth Naik	410972F		5		4	1	1		1	12/02/2013	2	4		2		2	nil		2	na		na	1	15			nil		nil		8.4
Gautam	388113F		9		6	1	1	na		11/01/2013	2	7		3		6	nil		2	na		na	1	20			nil		nil		9.2
Dhanya S	973919C		4	na		2	1	na		07/02/2007	2	4		4		4		2	2	na		na	1	10			nil		nil		10
aman nair	649506c		8	na		1	2	n		25/10/2005	2	4		4		8		5	2	na		na	1	15			nil		nil		7.1
Prabu C	758508C		22		20	1	1		1	10/01/2006	2	1		12		22		5	2	na		na	1	25			nil		nil		9.9
Prakash kumar nayak	790685C		28		28	1	1	na		14/03/2006	2	1		3	na		nil		2	na		na	1	10			nil		nil		12.5
Sumeet kumar	724311C		13		9	1	1		1	10/10/2005	2	9		1		1	nil		2	na		na	1	15			nil		nil		8.2
samartha das	951351c		5		2	1	2		2	22/12/2006	2	1		5	na		nil		2	na		na	1	15			nil		nil		10.1
Pethiraj	954969C		30	na		1	1	na		11/01/2007	2	1		30	na			5	2	na		na	1	20			nil		nil		9.2
Thenmozhi	083969D		12		11	2	1		1	17/08/2007	2	1		5		11	nil		2	na		na	1	10			nil		nil		8.5
Naveen Raja	083842D		5	na		1	1	na		27/11/2007	1	1		5	na		nil		2	na		na	1	15			nil		nil		9.3
urmila devi	102874D		30	na		2	2	na		18/09/2007	2	9		21		30	nil		2	na		na	1	10			nil		nil		8.8
deepali	609413C		10		3	2	1		1	15/03/2005	1	1		3	na		nil		2	na		na	1	10			nil		nil		8.4
Jiwan kumar	733038C		21	na		1	2	na		22/11/2005	2	1		3		3		5	2	na		na	1	12			nil		nil		9.7
Kalyani	609411C		6		1	2	1		1	15/03/2005	1	9		1		1		2	1		3	nil	1	20			nil		nil		8.2

WBC a	Plt	Retic at	MCV-Dx	creat	Hb-F at Dx	HbS at D	Hb-6m	mcv-6m	creat-6m	Tx-last 6m	Painful crise	Painful Crises F	Vc crises frqu	ACS-LFL	date of last fi	Complia	Response at last f/u
.	.	.	.	.	(%)	(%)	.	.	.	.	.	(/yr)	.	.	.	1-good	.
.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	2-Poor c	.
.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.
11200	363000	4.2		75.2	0.6	18.3	76.7	11.3	na	nil	nil	nil	nil	nil	18/02/2014	1	1
8800	413000	10.99		61.5	1	4.7	86.6	6.7		0.9	2 nil	nil	nil	nil	14/03/2014	1	2
14200	302000	12.48		84	0.4	12.2	82.2	9.5	88.5	0.4 nil	nil	1	nil	nil	04/06/2014	1	1
na	na	na	na		0.5	18.3 na		9.9	na	nil	nil	nil	nil	nil	06/06/2012	1	1
13700	518000	8		77.4	0.5	11.7	83	6.7	na	nil	1	1	nil	nil	16/05/2014	1	2
12400	205000	20.18		81.6	0.5	23.3	71	10.3	na	nil	nil	1	nil	1	26/12/2014	1	2
15400	699000	8.2		70	na	6.1	81.1	9.6	na	nil	1	3	nil	nil	22/08/2014	1	2
16900	426000	4		64	0.5	10.6	61	8.8	62.5	na nil	nil	1	1	nil	26/09/2014	1	2
15700	242000	14.75		87	0.4	20.7	73.2	8.2	na	nil	1	1	1	nil	21/04/2014	1	2
24800	112000	13.48		89.3	0.5	11.4	82.4	8.5	105.6	na nil	nil	1	nil	nil	27/04/2010	1	2
10500	459000	6.25		81.4	0.5	16.6	77.3	10.1	na	nil	1	1	nil	nil	25/09/2014	1	2
11700	186000	7.2		95	0.8	18.3	76.5	9	na	nil	nil	nil	nil	nil	18/08/2014	1	1
8100	266000	6.4		96	0.6	29.7	66	10.8	na	nil	nil	0.25	nil	nil	28/03/2014	1	2
3400	89000	4.33		91.6	0.4 na	na		12	88.5	0.5 nil	nil	0.14	nil	nil	05/07/2014	1	2
6200	202000	5.87		69.1	0.5 na	na		11.1	na	6 nil	nil	nil	nil	nil	03/12/2013	1	3
28800	628000	12.5		91.7	0.4 na	na		8.1	111	na nil	nil	1	nil	nil	28/11/2014	1	2
14600	473000	4.26		76.8	0.5	22.3	73.4	11.1	85.5	na nil	nil	nil	nil	nil	28/02/2014	1	1
9400	225000	7.71		76.4	0.4	23.4	73.2	9.9	na	nil	nil	nil	nil	nil	02/06/2014	1	1
18500	562000	6		92.3	0.9	24.1	70.2	10.7	101.3	na nil	nil	nil	nil	nil	28/11/2014	1	1
15600	222000	11.68		77.7	0.6	26.1	62.2	8.8	na	1 nil	nil	nil	nil	nil	19/09/2014	1	2
9700	653000	2.99		94.9	0.6	25.9	68.7	11	na	nil	nil	nil	nil	nil	01/12/2014	1	1
3900	153000	na		84.8	0.6	24.2	73.2 na	na	na	nil	nil	nil	nil	nil	07/05/2014	1	1
17000	414000	18.5		100	0.5	12.6	83.3	9.6	na	nil	1	1	nil	nil	26/12/2014	1	2
8000	206000	3.39		83.6	1.1	29.5	64.4	7.8	85.4	1.1 nil	nil	nil	nil	nil	03/02/2011	1	1
10900	260000	3.99		64.5	0.5	25.7	67.7	8.4	72.3	0.6 nil	nil	nil	nil	nil	22/08/2014	1	1
12600	356000	13.2		72.3	0.5	30	59	11.1	71.4	na nil	nil	1	1	nil	03/06/2014	1	2
13300	171000	3.43		71.4	0.4 na	na		8.9	na	nil	nil	nil	nil	nil	07/02/2015	1	2
7700	195000	4.12		68.5	0.5	9.9	79.6 na	na	na	nil	nil	nil	nil	nil	12/09/2014	1	2
17700	783000	8.12		89.6	0.5	22.7	73	10.5	na	nil	nil	0.5	nil	nil	22/11/2014	1	2
9600	300000	5.49		88.7	0.6	24	71	10	na	nil	nil	1	nil	nil	03/02/2015	1	2
13500	592000	9		83.1	0.4	18.6	77.8	6.8	na	nil	nil	nil	nil	nil	01/08/2014	1	2
38400	445000	3.45		69.5	na	9	83.2	7.1	na	2 nil	nil	nil	1	nil	18/11/2014	1	2
9200	313000	6.2		69.6	0.5	30.6	53.4	8.9	na	nil	nil	nil	nil	nil	18/03/2014	1	1
19900	289000	na		77.8	0.7 na	na		12.5		0.6 nil	nil	0.33	nil	nil	22/01/2015	1	2
20400	390000	4.87		67.1	0.4	54	45.1	9	75.1	na nil	nil	nil	nil	nil	18/03/2014	2	1
19700	246000	9.36		91.8	0.4	16.9	79.1 na	na	na	nil	nil	nil	nil	nil	31/10/2014	1	1
6100	126000	5.31		74.4	0.5	23	71	10.7	na	nil	nil	0.25	nil	nil	12/12/2014	1	1
11600	180000	10.49		74.4	0.5	19.6	71.1	11.6	na	na nil	nil	1	nil	nil	19/08/2014	1	2
10800	401000	na		75.2	0.9	8.6	82.8 na	na	na	nil	nil	nil	nil	nil	15/09/2014	1	1
14700	362000	na		89.3	0.5	15.8	52.9	8.5	na	nil	nil	nil	nil	1	21/11/2014	1	2
13700	273000	4.32		74.2	0.7 na	na	na	na	na	nil	nil	nil	nil	nil	11/04/2014	1	1
11900	356000	8.14		81.1	0.5	25.8	69.9	11.3	na	nil	nil	nil	nil	nil	14/03/2014	1	1
7000	90000	7.58		96.1	0.9	23.1	74.5	11.6	na	nil	nil	nil	nil	nil	10/05/2013	1	1
9000	176000	6.7		99.3	1.3	20.2	75.8 na	na	na	na	na	nil	nil	nil	06/05/2014	2	3
14500	201000	6.38		59.5	na	6.1	81.3	7.8		0.5 nil	nil	nil	nil	nil	21/12/2012	1	1
14400	446000	7.44		94.3	na	17.4	77.2 na	na	na	na	nil	nil	nil	nil	12/03/2014	1	1
12100	108000	5.95		70.1	na	25.4	65.4	9.7	na	nil	nil	nil	nil	nil	02/09/2014	1	1
7400	236000	2.43		72.5	0.5	11.6	55.3 na	na	na	1 nil	nil	nil	nil	nil	05/09/2014	2	1
10600	526000	6.29		63.5	1	9.6	79.9	9.6	81.3	na nil	nil	2	nil	nil	17/09/2014	1	2
8200	146000	2.24		71.2	0.4	18.7	76.8	9.2	na	nil	nil	nil	nil	nil	13/01/2015	1	1
5900	191000	2		106	0.6	22.8	72 na	na	na	nil	nil	nil	nil	nil	26/11/2014	1	1
4500	102000	2.66		88.7	0.9	16.8	71.2 na	na	na	na	na	nil	nil	nil	05/09/2014	1	2
39600	392000	na	na		0.5	21.8	72	10	101.8	na nil	nil	nil	nil	nil	05/12/2014	2	1
5900	291000	na		68	0.4	12.7	82.2 na	na	na	nil	nil	2	nil	nil	03/06/2014	2	2
14900	535000	13.8		85.6	0.5	30.7	63.3	9.1	82.9	na nil	nil	nil	nil	nil	22/07/2014	1	1
15900	478000	2.67		78.1	0.7	16.9	71.8 na	na	na	nil	nil	1	nil	nil	23/08/2014	1	2

15400	141000	9.84	86.6	0.5	na		na		8.8		na		nil		nil			1	nil		nil		17/06/2014		1		2
5200	84000	7.71	81.3	0.7		22.4	52.4	na		na		na		nil		nil			1	nil		nil		13/01/2015		1		2
9800	213000	3.8	70.8	0.5	na		na		9		83.4	na		nil		nil		nil		nil		nil		20/11/2014		1		1
10000	250000	6.24	89.1	0.4	na		na		9.9		95.8	na		nil		nil		nil		nil		nil		05/08/2014		1		1
9200	345000	4.4	64.6	0.5	na		na		9.5		73.6	na			1		1	nil		nil		nil		30/10/2014		1		2
5400	253000	0.8	81.5	0.5	na		na		9.2		80.7	na		nil		nil		nil		nil		nil		10/02/2015		1		2
20600	511000	15.7	79.1	0.4	na		na		8.9			na			1	nil		nil		nil		nil		30/06/2014		1		2
10400	360000	7.74	90.8	0.6		11.4	83.8		8.7	na		na		nil		nil		nil		nil		nil		18/11/2014		1		1
13300	299000	9.7	101.7	0.5		24.1	69.4		11			na		nil		nil		nil			1	nil		09/09/2014		1		2
8700	179000	18.54	92.7	0.3		9	77.1		10.1		109.4		0.5		1	nil		nil		nil		nil		01/09/2014		1		1
7800	164000	4.99	72	0.79		28.8	62.4		10		75.9	na		nil		nil			0.5	nil		nil		09/01/2015		1		2
15900	337000	10.14	75.2	0.39		22.7	72.8	na		na		na		nil		nil			0.5	nil		nil		09/02/2015		1		2
6200	156000	5.65	70.4	na		16.2	78.2	na		na		na		nil		nil		nil		nil		nil		21/08/2014		1		1
18500	375000	8.47	92.9	0.66		16.6	79.2	na		na		na		na		na			0.3	nil			1	17/10/2014		2		2
18400	660000	3.7	67.4	0.8		12.5	58.2		9.9			na			1	nil			0.5	nil		nil		19/11/2014		1		2
10700	197000	4	57.7	0.5		19	69.9		11		82.9		0.7	nil		nil		nil		nil		nil		03/02/2015		1		1
14600	221000	3.89	63	0.5		29.6	58		8.8		75.5		0.5	nil		nil		nil		nil		nil		18/11/2014		1		1
5500	44000	1.92	67.6	0.82		12.2	76.3	na		na		na		nil		nil			1	nil		nil		13/03/2014		1		2
30800	193000	7.7	94.1	0.7	na		64		9.2		102.3		0.7	nil		nil		nil		nil		nil		22/12/2014		1		1
10700	234000	8.6	88.9	0.6		13.2	82		9.8		135.9		0.9	nil		nil		nil		nil		nil		12/01/2015		1		1
8100	81000	10.45	90.3	1		7.9	86.6		11.2		107	na		nil		nil		nil		nil		nil		04/08/2014		1		1
6700	114000	17.35	79.7	0.3		18.1	72.1		7.3		86.4	na			1	nil		nil		nil		nil		04/11/2014		1		2
10800	145000	11.62	100.6	0.32		22.1	66.3		7.8			na		nil		nil		nil		nil		nil		02/12/2014		1	na	
8500	196000	5.5	91	0.4		21.6	61.2		10.1			na		nil		nil		nil		nil		nil		08/04/2014		1		1
11800	448000	10	92.4	0.5		16	77.8		8.9		108.8	na		nil		nil		nil		nil		nil		06/01/2015		2		2
7500	153000	1.82	76	0.9		17.7	76.8	na		na		na		nil		nil		nil		nil		nil		05/01/2015		1	na	
8700	312000	0.87	86.7	0.5	na		na		11.6		97.6	na		nil		nil		nil		nil		nil		22/12/2014		1		1
17100	349000	5.37	64.1	0.5		23	64.5		9.2		67	na		nil		nil		nil		nil		nil		28/11/2014		1		2
6800	163000	0.94	82.1	0.43	na		na		10		88.3		0.3		1	nil		nil		nil		nil		09/01/2015		1		1
9900	175000	7.5	62.7	0.4	na		na		8.5		73.2	na		nil		nil		nil		nil		nil		17/06/2014		1		1
9100	281000	6.7	77.9	0.5		33.3	61		9.3		87.1	na		nil		nil		nil		nil		nil		25/03/2014		1		1
15800	297000	5.9	63.4	0.2	na		na		9.4		72.8	na			1	nil		nil		nil		nil		08/12/2014		1		1
14600	268000	3.12	74.5	0.4	na		na		10.6		93.6	na		nil		nil		nil		nil		nil		20/02/2015		1		1
5000	17000	8.15	79	0.8		16.1	38.2		7.7		86.5	na			8		1		1	nil		nil		09/05/2014		1		3
3600	96000	12.6	73.6	0.5	na		na		5.5				0.7		6	nil		nil		nil		nil		06/02/2015		1		1
10400	206000	12.2	82.3	0.4		25.7	74	na		na		na		nil		nil		nil		nil		nil		29/01/2015		1		1
14700	271000	10.6	75.4	0.57		14	80.5		9.2		80.9	na			3	nil		nil		nil		nil		26/12/2014		1		2
8700	126000	5.12	93.8	0.7		21	74.8	na		na		na		nil		nil		nil		nil		nil		03/02/2015		1		1
9900	202000	7.99	99.2	0.8		21.3	71.9		9.4		109.7	na		nil		nil		nil		nil		nil		26/06/2014		1		1
9200	120000	3.58	74.2	0.86		20.5	72.1	na		na		na		nil			2		2	nil		nil		17/05/2014		1		2
16000	302000	10.77	92.1	0.3		24.5	71.7		10.4		96.5	na		nil		nil		nil		nil		nil		27/10/2014		1		1
22700	160000	18.3	88.9	0.75		25.3	70.8		9.8		89.4	na		nil		nil		nil		nil		nil		22/12/2014		1		1
6900	108000	13.95	66.8	0.4		24.2	70.5		8.9			na		nil		nil		nil		nil		nil		13/10/2014		1	NA	
17000	352000	6.28	88.8	0.6		19.5	73.8		10.5			na		nil		nil		nil		nil		nil		09/05/2014		1		1
22100	332000	3.22	71.8	0.5		36.6	55.6		11.6		83.5	na		nil		nil		nil		nil		nil		30/12/2014		1		1
13200	397000	9.19	74.2	0.38		17	77.8		6.5		87	na			1	nil		nil		nil		nil		31/03/2014		1		2
7800	107000	3.3	73.5	0.7		19.9	69.9		10.4		72.8	na		nil			1		0.5	nil		nil		28/07/2014		1		2
9300	278000	2.6	69.5	0.7		18	75.7		10.1		73		0.3	nil		nil		nil		nil		nil		12/08/2014		1		1
6000	117000	11.53	88.9	0.3		9.5	51		10.8		108.8		0.3		1	nil		nil		nil		nil		02/12/2014		1		1
7300	82000	2.17	86.3	0.5		41.9	27.8	na		na		na			1	nil		nil		nil		nil		08/07/2014		1	NA	
10400	221000	7.98	85.9	0.8		7	86.4		8.9		117.5	na		nil		nil		nil		nil		nil		08/09/2014		2		1
11500	392000	11.3	104.7	0.8		23.4	71.2		7.7		118	na			1	nil		nil		nil		nil		21/10/2014		2		2
16500	395000	11.89	87.4	0.4		22	73	na		na		na		nil		nil		nil		nil		nil		15/07/2014		1		1
7800	191000	6.38	81.1	0.76		25.7	69.3	na		na		na		na		na		nil		nil		nil		12/08/2014		1		1
13900	192000	5.39	82.7	0.8		22.2	68.7		11.2		75.4	na		nil		nil		nil		nil		nil		17/02/2015		1		1
10800	181000	4.3	72	0.6		10.4	83.7		10.5		76.5	na		nil		nil			1	nil		nil		18/07/2014		1		2
8500	124000	4.7	63.2	0.5		19.5	73.1		11.3		81.9	na		nil		nil		nil		nil		nil		16/01/2015		1		1
5400	113000	13.4	95.3	na		21.9	73.4	na		na		na		nil		nil		nil		nil		nil		02/12/2014		1		1
6700	214000	2.68	75.5	na		23	70.4		12.7		83.5	na		nil		nil		nil		nil		nil		23/12/2014		1		1
17000	538000	6.5	77.3	0.5		19.5	73		9.7		84.5		0.3	nil		nil		nil		nil		nil		09/09/2014		1		1

8000	279000	4.3	75.6	0.4	20.1	71.1	10.6	81.1	0.4	nil	nil	nil	nil	nil	09/09/2014	1	1
13700	378000	7	78.9	0.9	14.9	80.2	na	na	na	nil	2	2	nil	nil	26/09/2014	2	2
18700	213000	na	88	0.5	7.6	87.3	10.6	na	nil	nil	nil	nil	nil	08/09/2014	1	1
11300	355000	5.13	101.5	0.5	24.6	70.1	na	na	na	nil	nil	nil	nil	nil	12/08/2014	1	1
10100	279000	2.79	67.6	0.6	16.9	76.7	na	na	na	nil	nil	nil	nil	nil	11/08/2014	1	1
18200	407000	8.8	87.2	na	25	69.7	na	na	na	nil	1	nil	nil	nil	10/06/2014	1	1
10200	357000	5	92.3	0.5	19.1	71.7	10.1	111.4	na	nil	nil	nil	nil	nil	17/06/2014	1	1
13500	309000	6.2	76.3	0.5	21	74.1	9.4	na	nil	nil	nil	nil	nil	10/06/2014	1	1
12100	426000	6.15	79.3	0.2	16.8	77.9	9.5	na	nil	2	2	nil	nil	09/09/2014	2	2
10200	161000	7.32	75.1	0.5	12.1	82.6	8.7	na	nil	nil	nil	nil	nil	09/09/2014	1	1
6700	77000	5.9	98.7	na	na	na	12.1	118.1	na	nil	nil	nil	nil	nil	12/12/2014	1	1
15200	228000	6.2	66.1	0.5	na	na	7.7	67.4	na	nil	nil	nil	nil	nil	16/12/2014	1	1
6200	235000	3.3	69.2	0.6	16.8	70.2	na	na	na	nil	nil	nil	nil	nil	23/05/2014	1	1
10400	513000	3.3	67.5	0.5	11.4	43.4	na	na	na	nil	nil	nil	nil	nil	10/06/2014	1	1
10400	257000	4.7	68	0.8	na	na	na	na	na	nil	nil	nil	nil	nil	30/09/2014	1	1
13000	332000	5.1	85.7	0.3	20.2	75.4	na	na	na	nil	nil	nil	nil	nil	10/10/2014	1	1
9200	101000	18.8	89.5	0.4	na	na	8.7	80.3	0.5	nil	nil	1	nil	nil	08/11/2011	2	2
14200	396000	4.4	na	0.4	17.6	69	9.8	83.3	na	nil	nil	nil	nil	nil	28/06/2011	2	2
10500	166000	8.3	78.5	0.7	na	na	11	98	na	nil	nil	1	nil	nil	09/07/2010	2	2
6500	118000	8.4	90.1	na	17.7	76.8	na	na	na	nil	nil	nil	nil	nil	11/07/2009	1	1
7300	154000	13.16	105	0.5	21.8	73.4	8.6	na	nil	nil	nil	nil	nil	06/01/2015	2	1
15900	416000	7.4	83.2	na	17.8	77.8	10	na	nil	nil	1	nil	nil	07/09/2009	1	2
20100	163000	23.6	108.2	0.4	18.2	68.1	na	na	na	nil	nil	nil	nil	nil	26/07/2011	1	1
15300	302000	7.7	75.1	0.6	13.4	81	na	na	na	nil	1	1	nil	nil	08/12/2008	2	2
5400	132000	4.12	87.7	na	33.3	65.2	9.6	95.2	na	nil	nil	1	nil	nil	01/12/2008	1	2
11200	289000	2.3	82.3	0.4	12	na	7.4	87.8	na	nil	nil	nil	nil	nil	08/10/2008	1	1
5600	175000	2.5	70.4	0.8	25.3	64.4	na	na	na	nil	1	1	nil	nil	05/07/2013	1	NA
15000	374000	4.2	86	0.7	17	76.9	na	na	na	nil	1	0.5	nil	nil	04/05/2012	2	2
17000	365000	10.3	89.9	0.4	28.1	70	10	na	nil	nil	nil	nil	nil	26/08/2008	1	1
11500	255000	2	84.9	0.5	19.4	64.4	9.8	97.9	na	nil	nil	nil	nil	01/04/2011	1	1
12000	405000	5.2	98.6	0.7	20.4	73.7	10.2	0.8	nil	nil	nil	nil	nil	13/10/2011	1	1
15600	400000	na	94.1	0.6	17.3	77.2	8.5	na	nil	nil	nil	nil	nil	26/02/2011	1	1
8000	323000	6.1	65.7	0.4	na	na	na	na	nil	nil	nil	nil	nil	24/12/2013	1	1
18200	503000	12.9	95.1	0.7	18	80	7.6	na	nil	nil	nil	nil	nil	15/10/2011	2	2
28600	499000	7.4	79.3	0.5	na	na	10.6	102.5	na	nil	nil	nil	1	nil	05/03/2013	1	2
13800	136000	4.86	65.8	0.5	na	na	9.4	76.3	na	2	nil	nil	nil	nil	23/04/2012	1	2
9800	114000	4	67.1	0.6	na	na	na	na	nil	nil	nil	nil	nil	nil	22/02/2013	1	NA
6200	120000	5.7	63.8	0.5	na	na	8.3	68.4	na	nil	nil	nil	nil	nil	29/03/2011	1	2
4900	189000	6.9	87.1	0.8	35	62	na	na	na	nil	nil	nil	nil	nil	19/02/2010	1	1
14300	329000	5.5	66.5	0.5	16	75	10.5	67.7	0.5	nil	nil	nil	nil	nil	16/07/2012	1	1
10100	212000	6.3	72.7	0.7	na	na	12.8	0.8	nil	nil	nil	nil	nil	nil	15/03/2013	1	1
8700	318000	5.3	94.8	0.7	19.9	75.8	na	na	na	nil	nil	nil	nil	nil	20/10/2011	1	1
12600	296000	11.5	89.9	0.6	12	83.7	7	na	nil	nil	nil	nil	nil	nil	23/09/2013	1	1
8000	253000	3.9	85.3	0.7	24.4	72.2	na	na	na	nil	nil	nil	nil	nil	22/06/2010	1	1
8100	70000	7.1	67.7	0.5	19	72.2	na	na	na	nil	nil	1	nil	nil	26/06/2013	2	2
13900	409000	5.6	91.5	0.5	27	66	na	na	0.6	nil	nil	nil	nil	nil	11/08/2011	2	1
14400	154000	8.2	97.2	na	na	na	na	na	nil	nil	nil	nil	nil	nil	31/07/2010	1	1
8100	362000	0.68	71.2	0.5	na	na	9.2	69.3	na	1	nil	nil	nil	nil	11/09/2009	2	1
8400	163000	4.6	65	na	35.5	55.6	8.4	65.8	na	nil	nil	nil	nil	nil	24/01/2009	1	NA
15600	276000	15.13	74.6	0.5	na	na	7.9	78.3	na	nil	nil	nil	nil	nil	21/10/2009	1	1
7300	116000	5.5	93.4	0.7	25.1	70.7	11	0.7	nil	nil	nil	nil	nil	nil	05/10/2010	1	1
20400	289000	5.34	66.1	0.5	11.3	77.5	na	na	na	nil	nil	nil	nil	nil	07/05/2013	2	1
9400	159000	3.94	90.8	0.6	18.2	75.2	na	na	na	nil	1	1	nil	nil	17/11/2010	1	2
13900	357000	10.15	93.5	0.6	12.7	80.6	na	na	na	nil	nil	1	nil	nil	30/05/2011	2	2
6000	54000	2.71	75.1	0.6	21.6	69.7	na	na	na	n	nil	nil	nil	nil	10/05/2011	2	1
15300	413000	3.9	95.5	0.7	18	55	na	na	na	nil	nil	nil	nil	1	10/02/2012	2	2
7400	240000	2.89	79.6	0.7	22	73.4	9.9	83.7	0.7	nil	nil	nil	nil	nil	14/02/2011	1	1
10000	3449000	5.9	62.4	0.4	11.6	77	na	na	1	nil	nil	nil	nil	19/06/2012	2	2
13400	319000	3.06	75.9	0.7	10.9	82.9	na	na	na	nil	nil	1	nil	nil	27/02/2015	2	2
5200	275000	7.51	84.6	0.7	27.8	68	10.4	88.6	na	nil	nil	nil	nil	nil	09/02/2013	1	1

12600	287000	5.63	73.4	0.7	8.7	78	na	na	na	nil	nil	nil	nil	nil	08/06/2012	2	1
18600	441000	8.5	103.3	0.5	18.1	76.9	9.9	109.1	na	nil	nil	nil	nil	nil	14/11/2012	1	1
5200	78000	4.5	96.7	0.9	24.7	71.9	12.5	101	na	nil	nil	nil	nil	nil	02/03/2012	1	1
15100	212000	7.2	80.6	0.5	11.2	75	9.6	91.1	na		2	1	1	nil	nil	03/05/2011	1	2
6400	267000	na		0.7	na	na	na	na	na	nil	nil	nil	nil	nil	nil	08/12/2011	1	1
10200	132000	4.43	73.1	0.7	19.4	70.4	na	na	na		1	1	0.5	nil	nil	01/08/2013	1	2
15800	557000	6.1	80	0.8	7.2	80.3	9.5	70.7	0.7	nil	nil	nil	nil	nil	nil	24/09/2013	1	1
12100	56400	7.4		0.5	25.2	73.1	na	na	na	nil	nil		1	nil	nil	10/04/2012	2	NA
8100	117000	5.06	66.6	0.8	19.9	70.6	na	na	na	nil	nil	nil		1	nil	19/12/2013	1	2
10000	376000	3.91	99.6	0.5	18	71.4	na	na	na	nil	nil	nil	nil	nil	nil	01/09/2011	1	1
26000	373000	5.85	71.1	0.3	16.7	71.7	6.8	68.8	0.4		1	nil	nil	nil	nil	19/06/2012	1	2
6500	73000	5.63	83.9	0.7	18.3	74.5	12.2	na	nil		1	nil	nil	nil	nil	31/12/2013	1	1
7500	310000	5.58	79.6	0.6	20.1	75.4	na	na	na	nil	nil	nil	nil	nil	nil	17/01/2014	2	2
13800	183000	23.48	92.6	0.6	26.3	70.4	8.9	95.1	na	nil	nil	nil	nil	nil	nil	06/08/2013	1	1
11500	529000	8.61	84.6	0.5	21.7	73.9	9.3		0.3	nil	nil	nil	nil	nil	nil	26/12/2013	1	1
11800	99000	7.21	69.3	0.5	13.6	74	11.3		0.7	nil	nil	nil	nil	nil	nil	20/08/2013	1	1
16900	181000	5.13	62.3	0.6	25.9	72.4	9.9	88.4	na	nil	nil	nil	nil	nil	nil	22/07/2013	1	1
10900	243000	3.59	80.2	0.4	23	64	na	na	na	nil	nil	nil	nil	nil	nil	30/04/2012	1	1
8800	112000	3.2	74.2	0.7	19.7	77.2	na	na	na	nil		1	1	nil	nil	04/02/2014	2	2
4900	56000	8.15	70.9	0.8	16.6	71	8.3		na		2	nil	nil	nil	nil	28/03/2012	1	2
5700	155000	8.47	75.2	0.5	na	na	7.4	82.5	na		4	nil	nil	nil	nil	15/01/2013	2	3
3900	152000	5.75	86.4	0.5	20.4	71.1	9.4	97	na	nil		1	nil	nil	nil	13/11/2012	2	1
16300	349000	1.87	84.7	0.5	na	na	10.7	92.4	na	nil	nil	nil	nil	nil	nil	08/06/2012	1	1
2100	95000	4.26	78.8	0.6	na	na	na	na	na	nil	nil	nil	nil	nil	nil	28/03/2013	1	1
10100	317000	6.8	59.9	0.4	30.1	64.6	9.6		0.4	nil	nil	nil	nil	nil	nil	25/02/2013	1	1
4400	113000	5.2	69.1	0.5	21	72.2	7.7	77.6	na	nil	nil	nil	nil	nil	nil	21/09/2013	1	1
8900	141000	4.8	63.7	1.1	14.7	68.4	9.5	86.3	na	nil	nil	nil	nil	nil	nil	17/06/2013	1	1
14000	346000	na	93.2	0.8	9.7	82.7	9.9	na	nil	nil	nil	nil	nil	nil	nil	26/12/2013	1	1
4500	65000	5.88	78	0.6	23.5	63.5	11.9	na	nil	nil	nil	nil	nil	nil	nil	03/12/2013	1	1
11600	224000	6.4	94.6	1.01	14.8	79.6	11.5	145.8	na	nil	nil	nil	nil	nil	nil	18/12/2013	1	1
4800	11000	5.6	82.2	1	na	na	na	na			4	nil		1	nil	19/04/2013	1	2
7800	12900	3.3	71.5	0.7	na	na	na	na	nil	nil	nil	nil	nil	nil	nil	13/02/2013	1	1
12400	120000	2.4	64.5	0.59	15.6	46	na	na	nil	nil	nil	nil	nil	nil	nil	12/11/2013	1	1
6000	175000	2.9	74.2	0.4	28	61	10.3	77.3	na	nil	nil	nil	nil	nil	nil	26/03/2013	1	1
8500	83000	9.3	88.8	0.89	na	na	11.5	na	0.8	nil	nil	nil	nil	nil	nil	03/10/2013	1	1
5600	99000	8.8	73.7	0.6	na	na	9.3	80.2	na	nil	nil	nil	nil	nil	nil	24/09/2013	1	1
6200	219000	8	86.4	0.4	na	na	12.2	na	na	nil	nil	nil	nil	nil	nil	10/10/2013	1	1
9900	215000	0.98	76.8	0.5	na	na	9.9	83.6	na	nil	nil	nil	nil	nil	nil	10/10/2013	1	1
11700	236000	6.2	71.8	0.6	na	na	9.2	66.9	na	nil	nil		1	nil	nil	01/10/2013	1	2
7000	211000	13.3	91.2	0.25	21.9	76.5	8.5	92.4	na	nil	nil	nil	nil	nil	nil	30/01/2014	1	1
17900	158000	9.2	86.3	0.5	na	na	na	na	na	nil	nil	nil	nil	nil	nil	24/10/2013	1	1
7500	297000	6.2	79.2	0.4	29.9	61.5	10.1	na	na	nil	nil	nil	nil	nil	nil	17/06/2014	1	2
10100	142000	23.76	72.1	na	24.8	65.6	8.7	na	na	nil	nil		1	nil	nil	09/06/2015	2	2
15300	398000	5.1	86.5	0.8	9.9	65	11.3	96.6	0.7	nil		1	1	nil	nil	18/07/2014	2	2
8900	193000	5.5	85.5	0.7	26.4	69.8	11.5	92.2	na	nil		1	1	nil	nil	24/06/2014	1	2
17400	118000	18.49	85.7	0.5	na	na	na	na			1	nil		1	nil	20/10/2014	1	2
9100	333000	3.37	73.7	0.5	29.2	58.4	na	na	nil	nil	nil			1	nil	15/12/2014	2	2
14000	na	na	na	na	7.5	66.1	na	na	na	nil	nil			1	nil	23/02/2009	2	2
15600	523000	11.8	96.7	0.5	na	na	10.9	114.6	na	nil		1	1	nil	nil	26/08/2011	1	2
13200	270000	4	85.5	na	27.3	68	na	na	na	nil	nil		0.25	nil	nil	06/12/2011	1	2
4800	82000	3.78	87.2	0.6	30.7	62.2	9.4	78.4	0.6	nil	nil	nil	nil	nil	nil	12/06/2012	2	1
13100	238000	9.2	83.5	0.4	12.2	81.8	8.4	88.4	na	nil		1	1	nil	nil	14/04/2008	1	2
8000	233000	4.8	68.4	0.6	12.1	78.8	9.3	na	0.6	nil	nil	nil	nil	nil	nil	08/03/2007	1	1
10100	148000	12.68	87.7	0.4	na	na	8.5	85.8	na		2	nil	nil	nil	nil	19/05/2010	1	3