Epidemiology of Sickle Hemoglobin in India

Ajit C. Gorakshakar

Abstract

The inherited disorders of hemoglobin are the commonest monogenic disorders in India. Of these, α-thalassemia is seen practically in every community of the country with varying frequency while sickle gene is confined mainly to dravidians and pre-dravidians tribes inhabiting malaria endemic regions. It is also seen among certain caste groups from coastal areas of Orrisa and Andhra Pradesh. Sickle hemoglobin was first detected by Lehman and Cutbush in 1952 among the tribals from Nilgiris. During the last 54 years, several groups of investigators conducted hospital based or epidemiological surveys in various ethnic groups. Based on these surveys, prevalence of sickle gene is found to be 0-18% in north eastern India, 0-33.5% in western India, 22.5-44.4% in central India and 1-40% in southern India and the gene frequency of Hb-S varies between 0.031- 0.41. Wide variability in the prevalence of Hb-S trait is observed in population groups within small geographical areas. Apart from malaria, factors like endogamy, ethnicity and inbreeding are responsible for this variability. Hence, a systematic study in a small geographic area like a district is required to understand the population structure and natural history of sickle syndromes. This should be followed by creating awareness, genetic counseling prenatal diagnosis (if possible) to control the birth of sicklers.

Normal Hemoglobin

Hemoglobin is the pigment present in the red cells that transfers oxygen to the tissues. It is a tetramer consisting of two pairs of non-identical globin polypeptide chains, each chain being associated with one heme group. It changes its structure during human development. In the fetus, the major hemoglobin component is HbF ($\alpha 2\gamma$ 2) while HbA ($\alpha 2\beta 2$) is a minor component. In adults, two components viz. HbA and HbA_2 ($\alpha 2\delta 2$) exist. HbF is also present in small quantities. In all Hb components two a chains combine with two non a chains. Each of these chains is controlled by distinct genes. Genes for a chains are present on chromosome 16 while genes for all the non a chains are present on chromosome 11 in a specific manner collectively known as 'β-globin cluster' (Weatherall & Clegg 2001).

Inherited hemoglobin disorders

These disorders fall in two main groups.

a) The structural hemoglobin variants

b) The thalassemias which are caused by defective globin chain production.

Both these groups follow a recessive form of inheritance. The structural variants generally result from single amino acid substitutions in the α or β chains. Most of known structural variants are harmless but in some cases they may alter the stability or functional properties of the hemoglobin and lead to a clinical manifestation.

In India, three main structural variants are seen. They are HbS (Codon 6A>T), HbE (Codon 26 G>A) and HbD (Codon 121 G>C). Of these, HbS is mainly seen in scheduled tribes and scheduled castes of Central, Southern and Western parts of our country while HbE is confined to the Eastern or North Eastern region of India. HbD is seen in the populations of Punjab, Gujarat, Maharashtra (Balgir 1996). b-thalassemia is seen practically in every population group in our country

Pathophysiology of Sickle Hemoglobin

HbS has a substitution of valine for glutamic acid at the sixth position of the beta globin chain. Valine is a hydrophobic amino acid while glutamic acid is a hydrophilic amino acid. So this substitution creates a hydrophobic spot on the outside of the protein structure that sticks to the hydrophobic region of an adjacent hemoglobin molecule's beta chain. This clumping together or polymerization of HbS molecules into rigid fibres causes the 'sickling'of red blood corpuscles (RBCs). This results in deoxygenated RBCs. On oxygenation depolymerization takes place. Cycling between polymerization and depolymerization causes the RBC membrane to become rigid. These rigid, distorted RBCs cause blockage of small blood vessels.

Those individuals with single globin chain defect are called carriers or traits or heterozygotes and they are generally symptomless. When the gene for sickle hemoglobin is inherited from both parents, it is called sickle cell anemia. As mentioned earlier, HbD, HbE and β-thalassemia genes are prevalent in various population groups in our country. Therefore, the gene for sickle hemoglobin may combine with these defective genes and form various genotypes like SD, SE, S β- thalassemia. These genotypes reveal variable clinical manifestations. The asymptomatic genotypes include sickle cell trait & the double heterozygous condition of HbS and Hereditary Persistence of Fetal Hemoglobin (HPFH). The symptomatic genotypes can be divided into mild or severe conditions. Hb S/δ β thal, HbS / β thal with mild mutation cause mild conditions, HbS / HbD show variable clinical severity while HbS / Hb E can be severe.

Prevalence of HbS

Sickle gene was first detected by Lehman and Cutbush (1952) among the tribals of Nilgiri Hills. Since then, more than 300 tribal groups have been screened to look for the presence of sickle gene (Bhatia & Rao 1987, Balgir 1996, Mohanty & Mukherjee 2002). The prevalence varies considerably among different tribal groups ranging from 0-35%. In certain states like Madhya Pradesh, Orissa, Chattisgarh, Jharkhand, Gujrat and Maharashtra it forms a major public health problem. It has also been detected among scheduled caste populations (Table 1).

Table 1: Prevalence of HbS in different scheduled caste populations

Caste	Place	HbS (%)	Reference
Mahars	Aurangabad	5.14	Lele et al 1962
Pana	Orissa	8.9	Ambekar et al 2001
Haddi	Orissa	6.8	Balgir 2005

Although it is considered as a 'tribal gene' it has been detected in certain general caste groups also (Table 2). Still certain population groups of various states remain untouched as far as screening for sickle gene is concerned, and data on the prevalence of Hb S is being published even today (Ramesh et al, 2006).

Table 2: Prevalence of HbS in general caste groups.

Caste	Place	HbS (%)	Reference
Brahmins	Nagpur	Few Cases	Gangakhedkar 1989
Muslims	"	"	"
Marathas	Pune	"	Ambekar et al 2001
Koshti	"	"	"
Khandyat	Orissa	20.7	Balgir 2005
Chasa	"	19.8	"
Kshatriya	"	6.8	"
Karan	"	4.2	"
Agharia	"	3.0	"

Magnitude of the problem

About 7% of the world's population is carriers of some form of hemoglobin disorder. There are about 270 million carriers of sickle cell anemia and/or thalassemia. (WHO 1994). In India, sickle cell gene is mainly restricted to tribal and scheduled caste population where carrier frequencies range between 5-40% or more with three focal points (Bhatia and Rao 1987). Based on the 1981 census and prevalence of Hb S in various populations studied, Rao (1988) estimated the expected number of sickle homozygotes as 1,31,375 in our country while expected number of sickle cell heterozygotes was 24,34,170. At that time the sickle cell gene was detected in 75 districts form various states. Kate (2000) compiled the data generated by various groups by screening various tribal populations from Maharashtra. This revealed that average prevalence of sickle cell carrier among the tribal population was 10% and that of homozygotes was 0.5%. Considering the tribal population of the state as 90 lakhs (census 1991), the expected carriers of sickle cell would be 9 lakhs and expected number of homozygotes would be 45000. Sickle gene is not seen frequently among the tribals of North East India with an exception to Panika from Mirajpur, Himalayan region as reported by Negi (1967). Sickle gene has also been reported among other caste groups (Balgir, 2005). In our country, tribals with sickle gene are mainly concentrated in Madhya Pradesh, Orissa, Chattisgarh, Jharkhand, Gujrat, Andhra Pradesh etc. Therefore as mentioned earlier, there are three focal points for sickle gene where this gene reached very high frequency.

Laboratory Diagnosis

Sickle cell gene is mainly prevalent in tribal populations generally living in remote areas. So screening has to be carried out in the field using minimum amount of blood. The test should be rapid simple, inexpensive yet reliable. Sickling test and solubility test are very simple and can be performed in the field but they can not be used for newborn screening. The other tests include cellulose acetate electrophoresis at pH 8.9, Iso-electric focusing and automated HPLC. These tests can be established at the main laboratory and may be used as confirmatory tests. Surve et al (2000) screened 3,246 samples from tribal

populations of the Dhule and Gadchiroli districts of Maharashtra by solubility test, Hb-electrophoresis and automated HPLC and showed that the overall sensitivity of solubility test is 93.8% and specificity is 100% so they concluded that positive results of solubility test should be confirmed by either Hb electrophoresis or HPLC. Similarly, if possible molecular techniques to detect HbS can also be established.

Hb electrophoresis followed by quantitation of various bands has an added advantage. Based on amount of Hb S present in each individual Rao & Bhatia (1988) could predict the prevalence of α-thalassemia. Using the same approach, Mukherjee et al (1998) also predicted the prevalence of α-thalassesmia in 124 sickle cell heterozygotes and confirmed it by Southern Blotting. So this technique seems to be a suitable method for screening for α - thalassemia in population studies. This is very important as it is one of the ameliorating factors in the clinical manifestation of sickle cell anemia (Mukherjee et al 1997).

Clinical Manifestations

Clinical picture of sickle cell disease in our country is extremely variable. Earlier it was believed that the course of this disease is benign but several reports from various parts of the country have shown that this is not true (Rath et al 1982; Kar and Devi 1997; and Sahu et al, 2003). Anemia, hepato-splenomegaly and vaso-occlusive crisis in the form of bone and joint pains and infections are very common. The other symptoms include jaundice, fever, gall stones and epistaxis priapism and leg ulcers are not common in India as reported by Mohanty and Mukherjee (2002).

Little information is available on the causes of death at different ages but several symptoms have been reported in sickle cell disease (SCD) cases from different parts of India. Sarma et al (1986) in 1812 cases of SCD have reported that there were 33 deaths (1.81%) due to SCD. The causes of mortality were septecemia, acute splenic sequestration, severe anemia and hemolytic crisis. Improvement in the life style of the tribals would help to control morbidity and mortality especially among sicklers. Extensive health education programmes must be initiated in these areas to create awareness about various symptoms of SCD especially in the pediatric age group. In a study from Central India, it has been shown that the prevalence of malnutrition and non-compliance to medications increased their susceptibility to infection (Patel, Athavale 2004).

Prevention programme

It is well known that sickle cell gene is widely prevalent in tribal populations. Although large number of population groups have been screened to find the prevalence of the gene still many are yet to be screened. They should be screened with the help of a competent hematology laboratory. Once the prevalence is established, awareness and understanding about the disease should be generated amongst them. The simplest way to prevent the birth of affected children is the avoidance of marriage between ‘trait’ persons. Early detection of SCD is extremely useful for improving the quality of life. Therefore neonatal screening for SCD is recommended. Once the cases are identified counselors should explain to the parents about general supportive measures like use of oral rehydration solution, drinking lot of water, avoiding adverse climatic conditions etc. complete immunization, prophylactic dose of penicillin and pneumococal vaccine may avoid repeated infections. It would help to study the natural history of the disease. Hence a cohort with long term follow up would generate important data on clinical

manifestation. But, with the current scenario, it is difficult to initiate neonatal screening among tribals in our country because the deliveries are generally conducted by elderly untrained ladies from their community.

Prenatal diagnosis is one of the important aspects of prevention programme. Colah et al (2005) offered prenatal diagnosis for sickle syndromes to 85 'at risk' couples from Maharashtra, Gujrat, Madhya Pradesh and Andhra Pradesh. They commented that there is a dilemma in counseling couples for prenatal diagnosis for sickle syndromes because if the course of the disease is mild, it is unnecessary to go for prenatal diagnosis.

Therefore for a successful prevention programme, good peripheral units performing reliable screening tests should be established. Awareness about the disorder should be generated. Screening should be conducted in all tribal population groups living in different geographical areas which are yet untouched and intervention programmes should be initiated in these areas to get detailed clinical picture of the disease.

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