Prevalence Of Haemoglobinopathies In Gujarat, India: A Cross-Sectional Study

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Citation

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Abstract

Various haemoglobinopathies are major public health problem in Gujarat, a state located in the western part of India. The data pertaining to their occurrence and prevalence in the state of Gujarat are scarce and hence it was considered worthwhile to study the burden of haemoglobinopathies in Gujarat, India. A retrospective analysis of blood samples of 428 cases referred to the pathology laboratory from various private practitioners/Government hospitals for the workup of anemia or other blood related disorders was done by Bio-Rad D-10 instrument. 153 (35.7%) patients out of 428 had haemoglobinopathies. Thalassaemia minor (70 cases, 16.35 %), thalassaemia major (32 cases, 7.48%), sickle cell disease (22 cases, 5.14 %) and sickle cell trait (12 cases, 2.8%) were most common haemoglobinopathies. Less prevalent haemoglobinopathies were sickle-?-thalassaemia, ?,?-thalassaemia heterozygote, Hb D trait, Hb E trait, Hb E- thalassaemia, Hb D disease, Hb E disease and sickle D disease. Our study indicates that almost all the common haemoglobinopathies are prevalent in Gujarat but sickle cell trait/anemia and ? thalassaemia are very common.

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INTRODUCTION

Haemoglobinopathies are a group of genetic disorders of haemoglobin (Hb). Haemoglobin is a complex molecule contained within erythrocytes that binds to and transports oxygen and carbon dioxide in the body. Defects in genes of haemoglobin can produce abnormal haemoglobins and anemia, which leads to conditions, termed as "haemoglobinopathies". Abnormal haemoglobins appear in one of two basic circumstances: decreased production of one of the globin chain e.g. thalassaemia, abnormal globin chain e.g. sickle cell disease [1,2,3].

-thalassaemia is a heterogeneous group of inherited disorder of haemoglobin synthesis, characterized by a reduction of ($^{+}$) or absence ( ) of synthesis of beta globin chains of haemoglobin. This results in an imbalanced chain synthesis, which determines the severity of the disease [4].

These hereditary disorders of haemoglobin pose a massive health problem in many countries including India [5]. The distribution of specific disorders varies geographically and by community [6]. WHO figures estimate that 5 % of the world population is carrier for haemoglobin disorders [7]. They cause moderate to severe hemolytic anemia leading to high degree of morbidity and mortality. The frequency of -thalassaemia in India ranges from 3.5 to 15 % in general population. Every year 10,000 children with thalassaemia major are born in India, which constitutes 10 % of the total numbers in the world [8]. The overall gene deletion frequency is 0.05 to 0.98 % but it is very high in India [9]. In west central Gujarat, it is as high as 95 % [6].

The average frequency of haemoglobin S (Hb S) is 4.3 % in India. The range varies from 0-44 %. It is 0-18.5% in northeast zone, 0-33.5 % in west zone, 22.5-44.4 % in the central zone and 1-40 % in the southern zone [10] . Sickle gene in India is mostly found amongst Dravidian and predravidian tribes [11] . Haemoglobin E (Hb E) is mostly present in the northeastern states of India [12] . Frequency of Hb E in Assam is 52 %, 7 % in Manipuris and 3.33% in West Bengal [9,13] . Hb E has also been documented in people from orissa, uttarpradesh, rajasthan, Bihar and Punjab [14,15] .

The frequency of Haemoglobin D (Hb D) has been reported to be 0.5 to 3.1% in different castes of Uttar Pradesh [16] . Hb D has also been reported from Bengal, Bihar, South India and Gujarat [17] .

The main objective of the study was to know the prevalence of haemoglobinopathies in the state of Gujarat, located in the western part of India and to review various strategies that could be implemented for the effective control and prevention of these disorders.

MATERIALS AND METHODS

The present cross-sectional retrospective study included 428 patients referred for screening of haemoglobin disorders from September 2005 to April 2006 at Green Cross Pathology and RIA Laboratory, Ahmedabad - a reference laboratory which received various samples for testing and diagnosis from many small laboratories and clinicians, from all over Gujarat. Hence, the blood samples were collected from the patients who visited Green Cross Laboratory or alternatively the samples were collected and sent by other pathology laboratory/clinicians from Civil Hospital, Ahmedabad; Shingala Laboratories, Jamnagar; Guru Pathology Laboratory, Palanpur etc. for testing of parameters like hemogram, peripheral smear, haemoglobin analysis by HPLC (Bio-rad D-10, Bio-Rad Laboratories, USA) and sickling test. Hemograms were done on automated 5-part differential cell counter (cell dyne, Abbott Laboratories, USA). Haemoglobin analysis was done on Bio-rad D-10 using -thalassaemia dual program. Haemoglobin analysis by Bio-rad D-10 is based on the principle of High Performance Liquid Chromatography (HPLC). Clinical history and physical findings were recorded as provided by the referring physician.

The peripheral smear was stained with Leishman's stain (Merck, India). Grading of hypochromia, anisocytosis, microcytosis, macrocytosis and polychromasia was done according to the standard criterion. Inclusion bodies (basophilic stippling), sickle cells, target cells, nucleated red cells, spherocytes and schizocytes were noted in peripheral smear, when seen.

RESULTS

153 patients out of 428 cases studied had haemoglobinopathy. The patients ≤ 18 years of age were considered as pediatric patients. An Age and Sex wise distribution of patients with haemoglobinopathies is described in Table I.

Majority of the patients studied were Gujarati (native residents of the state of Gujarat) by origin. However, our study consisted mainly of hospital based case reports, which cannot be regarded as representative of a community or population. Majority of the patients had -thalassaemia and sickle cell disease/trait. Five patients of sickle cell disease were diagnosed in adulthood although they were symptomatic from childhood.

Figure 1

Table I: Age and Sexwise distribution of Patients with different haemoglobinopathies

Haemoglobinopathies	Pediatric			Adult		
	Total	Male	Female	Total	Male	Female
Thalassaemia Major	29	20 (4.67%)	9 (2.10%)	3	3(0.70%)	0
Thalassaemia Minor	22	15 (3.50%)	7 (1.64%)	48	22 (5.14%)	26 (5.08%)
Sickle cell disease	17	14 (3.27%)	3 (0.70%)	5	2 (0.47%)	3 (0.70%)
Sickle cell trait	3	3 (0.70%)	0	9	4 (0.93%)	5 (1.17%)
Hb D disease	0	0	0	1	1 (0.23%)	0
Hb D trait	1	1 (0.23%)	0	1	0	1 (0.23%)
Hb E trait	1	1 (0.23%)	0	1	1 (0.23%)	0
Hb E disease	1	0	1 (0.23%)	0	0	0
Hb E + thalassaemia	2	1 (0.23%)	1 (0.23%)	0	0	0
Sickle thalassaemia	2	2 (0.47%)	0	1	0	1 (0.23%)
β–thalassaemia intermediate	2	1 (0.23%)	1 (0.23%)	0	0	0
δβ–thalassaemia	1	1 (0.23%)	0	2	2 (0.47%)	0
Sickle D disease	1	1 (0.23%)	0	0	0	0
No. of Patients	82	60 (14.01%)	22 (5.14%)	71	35 (8.17%)	36 (8.41%)
Total No. of Patients with Haemoglobinopathies	153 (35.75%)					
Normal Subjects in study population	275 (64.25%)					
Total No. of Patients studied	428 (100%)					

(No. of Patients with haemoglobinopathies = 153)

(Numbers in the parenthesis indicate the observed frequency (in %) of various disorders in the total numbers of samples studied (N=428)

13 patients out of 32 patients of thalassaemia major had

received blood transfusions before the investigations. Nearly 25.01% patients were diagnosed late as shown in Table II (15.63% between 3-10 years and 9.38 % > 10 years).

Figure 2

Table II: Age wise Distribution of Patients with Thalassaemia major (n=32)

Age	% of cases
≤ 3 years	75.0%
3 - 10 years	15.65 %
11 - 20 years	9.38 %

Table III describes blood indices and haemoglobin analysis of patients with abnormal haemoglobin studied by HPLC method.

Figure 3

Table III: Blood indices and Hb analysis results of common haemoglobinopathies

Laboratory Parameters	Thalassaemia Major	Thalassaemia Minor	Sickle Cell Disease	Sickle Cell Trait
Hb (gm %)	5.5 (1.14-13.5)	10.14 (3.2-14.1)	7.76 (2.31-12.1)	9.79 (5.05-13.9)
RBC (mill/cmm)	2.6 (0.57-6.01)	5.37 (1.35-6.65)	3.35 (1.14-5.77)	4.51 (1.9-5.55)
MCV (fl)	66.93 (55.7-80)	63.15 (48.1-83.1)	75.98 (58.8-92.8)	73.98 (46.8-95)
MCH (pg)	20.43 (12.4-30.1)	18.75 (10.1-27)	22.66 (17.5-28)	22.32 (110.8-30.1)
MCHC (%)	29.93 (21.6-38.7)	29.92 (23.5-35.8)	29.42 (24.7-34.2)	31.18 (23-36.4)
RDW (%)	30.59 (20.4-41.9)	15.04 (12-21.3)	19.59 (13.4-32.1)	16.43 (11.9-22.5)
HPLC - A2 (%)	2.96 (0.9-6.3)	6.09 (4.2-7.7)	3.51 (1.4-6.5)	3.58 (3-4.6)
F (%)	66.55 (14-85.6)	1.29 (0-8.8)	17.88 (4.7-30.2)	1.17 (0-4.2)
A (%)	14.95 (0.4-68.8)	81.33 (73.9-85.6)	5.89 (2.2-33.2)	55.83 (40.1-67.6)
S (%)			67.08 (38.6-80.6)	31.51 (20.3-47.5)

(Values in the parenthesis indicate the range of various parameters observed in the blood samples of patients with haemoglobinopathies)

Twenty-four patients (75%) with thalassaemia major had severe anemia at the time of diagnosis (< 7 gm % Hb). One patient had only 1.14 gm % of haemoglobin. Majority of the patients with sickle cell disease had blood indices and blood film suggestive of hypochromic microcytic anaemia (18 out of 22 patients, 81.8 %). One patient of sickle cell disease had received blood transfusion before investigations and hence had low Hb S (38.6 %) and high Hb A (33.2 %). Three patients (25 %) of sickle cell trait had severe anaemia (< 7 gm % Hb) due to associated problems. Six patients of sickle cell trait had blood indices and blood film suggestive of hypochromic microcytic anaemia. Seven patients (31.82 %) of sickle cell disease had very high Hb F (> 20 %). Eight patients (66.67 %) of sickle cell trait had very low Hb F (< 1 %). Six patients of sickle cell trait had < 30 % of sickle haemoglobin. One patient of sickle cell disease had Hb F < 5 % and one had Hb F < 10 %.

All the patients with thalassaemia minor had low Mean Corpuscular Haemoglobin (MCH) (< 27 pg/dl), only one had Mean Corpuscular Volume (MCV) > 77 fl and 3 patients had high Red Cell Distribution Width (RDW). Four patients of thalassaemia minor had severe anaemia (< 7 gm % Hb).

DISCUSSION

The incidence of -thalassaemia minor and major was 16.35 % and 7.48 % respectively in the present study. This incidence coincides with the previous reports [18].

The frequency of sickle cell disease is 5.14 % in our data. The average frequency in India is 4.3% [19].

Hb E disease is most frequently found in Eastern and far Eastern parts of India [13,20]. Hb E is not very common is Gujarat [6]. The incidence of Hb E was very low (0.23 %) in this study.

The incidence of Hb D disease was low (0.23 %) in the present study which coincides with the previous reports [21].

Many patients of thalassaemia major had received blood transfusions (13 patients out of 32, 40.6 %) before the diagnosis and many patients were more than 3 years of age at the time of diagnosis (25.01%). A thalassaemia intermedia is suspected when a patient presents after 3 years of age or needs fewer blood transfusions. Early splenectomy is helpful in thalassaemia intermedia. Some of our patients could have thalassaemia intermedia but since they were transfused before the diagnosis, it was difficult to differentiate between thalassaemia intermedia and thalassaemia major. Majority of the patients (75 %) of thalassaemia major had severe anaemia at the time of diagnosis indicating lack of awareness about the disease, in treating clinicians or tendency of the parents to seek advice of the doctors only as a last resort [22].

A patient with sickle cell disease or trait has normochromic normocytic anaemia but majority of the patients in this study had hypochromic microcytic anaemia (81.8 % and 50 % of

the patients with sickle cell disease and trait respectively) [23]. This could be due to associated iron deficiency or -thalassaemia trait. High incidence of iron deficiency has been reported in patients with sickle cell disease from India [21]. -thalassaemia trait should be suspected with sickle cell disease/trait when Hb F is unusually low with typical blood indices and low Hb S (< 30 %) [7,8,9]. One patient of sickle cell disease had Hb F < 5 % and one had Hb F < 10 %. Both had typical indices of thalassaemia trait. Eight patients (66.7 %) of sickle cell trait had very low Hb F (< 1 %) and six patients had low Hb S (< 30 %) [21,23,24]. Very high gene frequency of thalassaemia is reported previously from Gujarat. Sicklers from India have high Hb F giving protection from sickle crisis [25]. Normally Hb F is upto 20 % in sickle cell disease but 31.82 % of our patients had Hb F > 20 %. This could be due to associated hereditary persistant foetal haemoglobin (HPFH). To the best of our knowledge such high HB F in sickle cell disease is not reported from India.

Thus, haemoglobinopathies exert significant burden on India, especially in the western part of the country. Adequate measures and screening procedures should be adopted to reduce this burden. Screening is affordable and an accessible way to detect carriers, and can be offered in a range of settings in different societies: in high school, before marriage, or in antenatal clinics. Haemoglobinopathy testing should be performed concurrently with ferritin, serum iron and Total Iron Binding Capacity (TIBC) for:

- Pregnant woman with low red cell indices
- Pregnant woman from a high-risk ethnic background
- Partner of the pregnant woman should be tested at the same time as the pregnant woman
- Partners of individuals who are carriers for thalassaemia or a haemoglobin variant
- A family history of haemoglobinopathy or haemoglobinopathy carrier state
- Individuals from ethnic groups with a high prevalence of haemoglobinopathy
- Consanguinity

Effective prevention approaches to thalassaemia have now been demonstrated in many countries with diverse carrier-screening programmes. For example, in Cyprus, Greece, the Islamic Republic of Iran and Italy, premarital screening for thalassaemia is standard practice and national audit data are available; most at-risk couples are identified in time to be offered early diagnosis for the first pregnancy [26,27]. The majority of such couples use this service and produce healthy offspring. In the United Kingdom of Great Britain and Northern Ireland and other north-western European countries where prenatal diagnosis is generally available, screening is offered during pregnancy [7,28]. Besides, such screening programmes must be supported by public education and regulatory structures so that individuals may make informed decisions and that people are protected against discrimination as a consequence of their test results [29,30].

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