ORIGINAL ARTICLE

FREQUENCY OF β-THALASSEMIA TRAIT IN FAMILIES OF THALASSEMIA MAJOR PATIENTS, LAHORE

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Background: Thalassemia major is one of the most common genetic disorders in Pakistan and over five thousand new patients are added in the pool annually. This familial disease has both medical and social implications, and therefore there is a need to assess the magnitude of β-Thalassemia trait amongst family members of Thalassemia major patients. **Methods:** This cross-sectional descriptive study enrolled 674 blood samples from first degree relatives of registered patients of Thalassemia major at Sir Ganga Ram Hospital, Lahore. Peripheral blood smears were studied for abnormal morphology findings of microcytosis, hypochromia, poikilocytosis (tear drops, target cells) and Erythrocyte indices (haemoglobin, RBCs, mean corpuscular haemoglobin, mean corpuscular volume, mean corpuscular haemoglobin concentration) and Hb electrophoretic (HbA, HbA2, & HbF). **Results:** Hb electrophoresis showed 61% of the study subjects had haemoglobinopathies. Frequency of β-Thalassemia trait was highest followed by β-Thalassemia major, HbE trait, HbD Punjab and Hb intermedia. **Conclusion:** Findings strongly suggest screening for β-Thalassemia trait in families of Thalassemia major patients.

Keywords: Thalassemia Trait, Hb electrophoresis, Haemoglobinopathies

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INTRODUCTION

Diverse mutations in globin genes are major cause of thalassemias, which are considered major hereditary disorders worldwide.[1] β-thalassaemia is one of the most common single-gene inherited conditions in the world.[2] Almost 70,000 infants are born with β-thalassaemia worldwide each year, and 270 million people are carriers of haemoglobinopathies.[3,4] β-thalassemia is most commonly present among populations in all Mediterranean countries, as well as in Southeast Asia, India, Africa, Central America and the Middle East.[2]

No documentary register is available in Pakistan, but it is estimated that 5000-9000 children with β-thalassemia are born per year. About 9.8 million carriers are estimated in general population with a carrier rate of 5–7%.[5,6] In another study, β-Thalassemia carriers were estimated to be 8 million in Pakistan.[6] Modell and Darlinson, 2008 calculated national carrier frequency for β-thalassaemia of 4.6% in Pakistan.[7] Trends of consanguineous marriages, high fertility rate, high birth rate, low educational level, early marriages with unawareness has led Pakistan towards very high number of children with transfusion dependent Thalassemia in the world.[8] Week infrastructure of health provides very few screening facilities and cultural trends of cousin marriages of 40% has led towards high risk of congenital transmission of β-Thalassemia trait.[9] Adult haemoglobin is composed of two α and two β chains encoded by two α-globin genes on chromosome 16 and one β-globin gene on chromosome 11. β-Thalassemia is associated with two types of mutation β^0 and β^+. Persons with β-Thalassemia minor have one abnormal allele but if two abnormal alleles are inherited then result is β-Thalassemia major. Sometimes patients may present with milder symptoms in β-Thalassemia intermedia, though have both mutant alleles.[10] This study was designed to estimate the frequency and magnitude of β-Thalassemia in families of Thalassemia major patients so that magnitude of the problem can be assessed in target population of the country.

MATERIAL AND METHODS

In this cross sectional descriptive study, a total of 674 subjects were included in the study from January 2007 to March 2009. Blood samples of first degree relatives of Thalassemia Major patients registered at Ganga Ram Thalassemia centre Lahore were collected after informed consent. Samples were analysed at the Haematology department of Fatima Jinnah Medical College, Lahore.

Disposable syringes were used to collect 3ml of venous blood sample by ensuring standard aseptic procedure. EDTA (ethylene diamine tetra acetic acid) was used as an anticoagulant. Samples were immediately labelled and data entered into record registers. Peripheral cell counts and red blood cell indices (RBC, Hb%, HCT, MCV, MCH, and MCHC) were measured using standard procedure. Haemoglobin electrophoresis was carried out on agarose gel using the Beckman Coulter, Inc S.A. System with Paragon reagents. The resulting electropherograms were visually observed and evaluated on densitometer for level of various normal and abnormal Hbs. The collected data was analysed using SPSS-17.

RESULTS

Venous blood was collected from 674, 1st degree relatives of Thalassemia major patients as per predefined guidelines. Peripheral blood smears were prepared for all and Hb electrophoresis was performed on 441 (65.4%) samples because 233 (34.6%) subjects had normal peripheral picture in correlation to clinical symptoms so they were not included in electrophoresis. Table-1 shows the frequency of males and females in the study. A total of 46.1% of the population group under study was in child bearing age, which is the high risk for transmission of Thalassemia to the next generation.

Peripheral blood smear and RBC indices were recorded for all samples (Table-2). Hb electrophoresis was performed on samples with abnormal findings in peripheral smears, high RBC count and low MCV. Abnormal Hb patterns were found in 61%. β-Thalassemia trait was most common followed by β-Thalassemia major, β-Thalassemia intermedia, Haemoglobin D/S trait and least common was Haemoglobin E trait.

Table-3 shows mean value with standard deviations of various types of haemoglobins**.** HbA2 mean was 5.13 in β-Thalassemia trait as compared to lower value of HbA2 in normal patients. HbA2% mean was almost same for β-Thalassemia major and Hb D/S trait. Increase in foetal haemoglobin is indicator of abnormality but because of lower frequency of E, HbD Punjab and intermedia mean values are high.

Table-1: Age and sex-wise distribution

Age group (Yrs)	Male n (%)	Female n (%)	Total (%)
*0–15	169 (25.1)	155 (23)	324 (48.1)
16–45	139 (20.6)	177 (26.3)	316 (46.9)
46+	18 (2.7)	16 (2.3)	34 (5)
Total	326 (48.4)	348 (51.6)	674 (100)

*0=1–11 months of age

Table-2: Spectrum of haemoglobinopathies frequency on Hb electrophoresis

Findings	Male n (%)	Female n (%)	Total (%)
β-Thalassemia major	14 (3.2)	12 (2.7)	26 (5.9)
β-Thalassemia Intermedia	2 (0.5)	5 (1.1)	7 (1.6)
β-Thalassemia Trait	122 (27.7)	107 (24.3)	229 (52)
Haemoglobin D/S trait	1 (0.2)	4 (0.9)	5 (1.1)
Haemoglobin E trait	0 (0)	2 (0.5)	2 (0.5)
Normal	88 (19.9)	84 (19.0)	172 (38.9)
Total	227 (51.5)	214 (48.5)	441 (100)

Table-3: Mean value with standard deviation of various haemoglobins

Type of haemoglobinopathies	HbA%	HbA2%	HbF%
Normal	96.92±1.1	1.76±0.80	1.0
β-Thalassemia Trait	94.85±1.15	5.13±1.11	3.0±0.28
Haemoglobin E trait	37.20±58.60	32.05±9.12	30.7±43.48
β-Thalassemia Major	25.19±30.23	3.65±6.73	68.22±32.56
Haemoglobin D/S trait	31.42±31.92	3.28±2.9	64.8±29.44*
β-Thalassemia Intermedia	77.64±12.38	2.31±1.23	20.01±12.99

*(HbD/S) Hb F is replaced by HbD/S on Hb electrophoresis

DISCUSSION

Hb electrophoresis and DNA studies are gold standard for Thalassemia diagnosis though they are expensive, yet the burden of treatment with blood transfusions is far more than the expense of diagnostic technique.[11]

Affected persons with Thalassemia major are an indicator for transmission of different traits in high risk families. Study results show frequency of 61% for different types of Thalassemia disorders including 51.9% for β-Thalassemia trait. Ahmed *et al* reported lower percentage of 31 in screening for carriers in extended families of Thalassemia patients of Pakistani families.[9] Incidence of β-thalassaemia trait among the siblings was 58% in a study conducted in by Khattak *et al*, which is closer to our study results.[12] A study conducted in Karachi reported 62.2% siblings as β-Thalassemia carriers in immediate family members of the patients.[13]

Regional variations are seen in neighbouring countries. Most common haemoglobinopathies observed were β-thalassemia minor (21.3%) in Bangladesh.[14] In a study conducted in India, 57 out of 60 suspected cases on peripheral smears were found to have some type of Thalassemia trait.[1]

Exact data about the prevalence of haemoglobin disorders is not available in Pakistan but its vertical transmission can be prevented by proper screening and counselling in families of Thalassemia patients. Although spread of Thalassemia is difficult to prevent at this time in Pakistan because of unawareness, lack of education, remote health counselling facilities, but a program of health education, testing for the trait, genetic counselling, and easy accesses to prenatal diagnosis can provide families with full medical information to help them have healthy children. Young people need to learn about their carrier status early enough to consider all available options, including marriage and undertaking a pregnancy. An understanding of basic population genetics is required both for service planning and for correct interpretation of surveillance data.[15]

CONCLUSION

Screening for Thalassemia Traits in relatives of known cases of Thalassemia can reduce the burden of transfusion and burden of treatment on economy. Pre-marriage counselling particularly in cousin marriages can also help control Thalassemia.

Further research on frequencies and prevalence of different traits is required in establishing regional database. Also in resource constrained health setup peripheral blood smears can be studied in correlation to clinical findings and suspected cases can be diagnosed

with Hb electrophoresis techniques at well equipped Laboratories.

REFERENCES

1. Alwar V, Kavadia R, Singh N, Rameshkumar K. Hunt for hidden trait. J Lab Physicians 2009;1(1):15–8.
2. Cousens NE, Gaff CL, Metcalfe SA, Delatycki MB. Carrier screening for Beta-thalassaemia: a review of international practice. Eur J Hum Genet 2010;18:1077–83.
3. Abolghasemi H, Amid A, Zeinali S, Radfar MH, Eshghi P, Rahiminejad MS, *et al*. Thalassemia in Iran: epidemiology, prevention, and management. J Pediatr Hematol Oncol 2007;29:233–8.
4. Modell B, Khan M, Darlison M, King A, Layton M, Old J, *et al*. A national register for surveillance of inherited disorders: beta thalassaemia in the United Kingdom. Bull World Health Organ 2001;79:1006–13.
5. Ansari SH, Shamsi TS, Ashraf M, Bohray M, Farzana T, Khan MT, *et al*. Molecular epidemiology of β-thalassaemia in Pakistan: Far reaching implication. Int J Mol Epidemiol Genet 2011;2:403–8.
6. Black ML, Sinha S, Agarwal S, Colah R, Das R, Bellgard M, *et al*. A descriptive profile of β-thalassaemia mutations in India, Pakistan and Sri Lanka. J Community Genet 2010;1(3):149–57.
7. Modell B, Darlinson M. Global epidemiology of haemoglobin disorders and derived service indicators. Bull World Health Organ 2008;86:480–7.
8. Qurat-ul-Ain, Ahmad L, Hassan M, Rana SM, Jabeen F. Prevalence of β-thalassemic Patients Associated With Consanguinity and Anti-HCV - Antibody Positivity – A Cross Sectional Study. Pak J Zool 2011;43(1):29–36.
9. Ahmed S, Saleem M, Modell B, Petrou M. Screening extended families for genetic haemoglobin disorders in Pakistan. N Engl J Med 2002;347:1162–8.
10. Kumar V, Abbas AK, Aster JC. editors. Robbins Basic Pathology. Philadelphia: Elsevier Saunders; 2007.
11. Rosu M. One step forward in health promotion. J Med Life 2012;5:367–72.
12. Khattak I, Khattak ST, Khan J. Heterozygous beta thalassemia in parents of children with beta thalassemia major. Gomal J Med Sci 2006;4(2):52–6.
13. Ansari SH, Baig N, Shamsi TS, Saif-ur-Rehman, Ansari ZH, Behar Z, *et al*. Screening immediate family members for carrier identification and counselling: A cost-effective and practical approach. J Pak Med Assoc 2012;62:1314–7.
14. MesbahUddin M, Akteruzzaman S, Rahman T, Hasan AK, Shekhar HU. Pattern of *β*-Thalassemia and Other Haemoglobinopathies: A Cross-Sectional Study in Bangladesh. ISRN Hematology 2012;2012:659191.
15. Firdous N, Gibbons S, Modell B. Falling prevalence of beta-thalassaemia and eradication of malaria in the Maldives. J Community Genet 2011;2:173–89.

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