DOI: 10.7860/JCDR/2017/26068.10674

Case Report

Genetics Section

Prenatal Screening for Rare Co-Inheritance of HbE and β-Thalassaemia Traits in Western India

PARTH S SHAH[1], HARI SHANKAR P RAY[2], KETAN K VAGHASIA[3], SANDIP C SHAH[4], MANDAVA V RAO[5]

ABSTRACT

The mutations in Haemoglobin Beta (HBB) gene, bring about less or no production of Hb β-chain synthesis in affected cases, leading from minor to major types depending on haematological indices. In compound heterozygotic conditions, two traits are involved, in which one parent has HbE trait and the other has β-thalassaemia carrier (trait). Here, we report a family of Rajasthan, West India which had a proband (son) having HbE/ β-thalassaemia a co-inherited compound heterozygosity as revealed by DNA sequencing. It also contained upper levels of HbE with altered Hb and red cell indices showing asymptomatic to symptomatic state requiring blood transfusion periodically. The parents and Chorionic Villus Sampling (CVS) were HbE and β-thalassaemia traits only. Such case is rare in Western India and we recommend this family for genetic counseling and genetic testing before they want reproductive choices in future for better management in a society.

Keywords: Compound heterozygosity, Electrophoresis, Genetic testing, Phenotypic indices, Sequence analysis

CASE REPORT

A family in Rajasthan attended a clinic as the proband (son) confessed β-thalassaemia symptoms because of low Hb levels, altered Mean Corpuscular Volume (MCV) and Mean Corpuscular Haemoglobin (MCH) values as reported by clinician. Then this Rajasthani family was referred for genetic testing to our Research Laboratory Ahmedabad. They were asked to fill consent forms. They have two probands, i.e., son and CVS. The blood of trio samples and CVS were subjected to electrophoresis for Hb variants and other blood indices. In this study CVS was suggested by clinician as it is better technique detected earlier than amniocentesis, which helps to identify the abnormal genetic condition after 8-10 weeks of pregnancy. Eukaryotic DNA was extracted and was subjected to Sanger's DNA sequencer with appropriate primers. We detected that father had HbE trait (c.79 G>A) and mother was HBB: c.92 +5 G>C (β0) trait. However sequence analysis of proband revealed the son had combined c.79 G>A (HbE) and C 92 +5 G>C (β+) heterozygotic state, whereas CVS exhibited only c.79 G>A (HbE trait) mutation only. Phenotypic parameters indicated mild to moderate anemic conditions as suggested by a clinician, where HbE levels were high (91.3%) along with an altered red cell indices like MCV and MCH values followed by low Hb levels [Table/Fig-1 and 2].

DISCUSSION

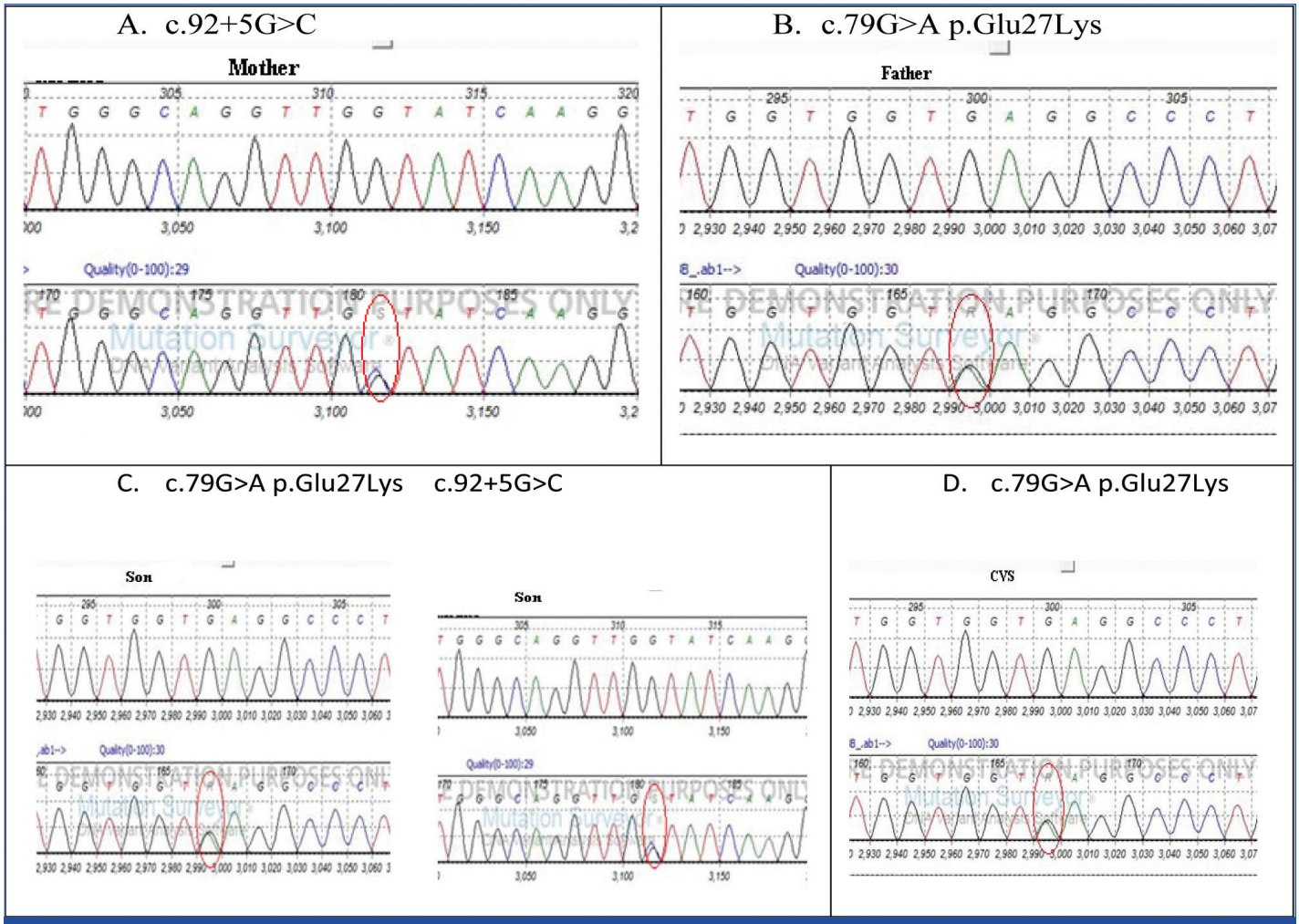
Thalassaemias of α and β types are blood borne genetic anomalies. These are autosomal recessive disorders inherited from one generation to other affecting children. The β-thalassaemia is dominant in Middle East, Mediterranean region, Asia, India, Africa and Far East [1-4]. About 3-17% of population is affected by β-Thalassaemia in Indian sub continent [5].

The β-thalassaemia minor (trait) co-exists with several other traits like HbD, HbE, HbS etc to become combined heterozygotic condition having minor, intermedia and major thalassaemia depending on phenotypic indices. The HbE/β-thalassaemia condition was more in North-Eastern Indian population and 50% of them have β-thalassaemia major. These patients presented a variable clinical futures presenting non-severe to severe anemic conditions requiring blood transfusions [6]. Agarwal S et al., detected high frequency of HbE/β-thalassaemia mutation (57%) in (21) families of North India [7]. Mondal and Mandal detected only 1.16% double heterozygous state in West Bengal [8]. But in Western India this condition is rare where, in a Gujarati family, co-inheritance of HbE/β-thalassaemia was reported with sickle cell anemia disease only [9]. In an exhaustive study documented by Vichinsky EP et al., HbE/β-thalassaemia predominated in Western America [2]. These variations could be due to several factors like interplay between qualitative and quantitative levels of Hb, Iron load, geographic distribution and gene polymorphism [7,10]. However, in our study also a rare condition of HbE/ β double heterozygous condition was detected in a Rajasthani family of Western India. The proband son was affected with such type of heterozygosity but the other CVS.

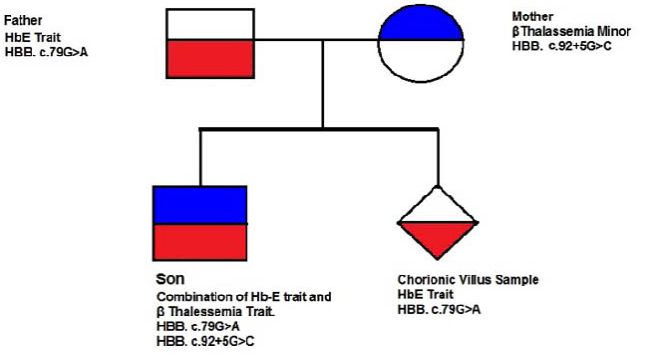
Sample exhibited only HbE trait. The son had this transmission, i.e., HBB c.79 G>A (HbE) and HBB c.92 +5 G>C (β+) from father and mother respectively and may require periodic blood transfusion

Sample	Haemoglobin indices	Units	Normal Range	Mutations	Genotype	Inference/ Clinical Report
Mother (35)	Hb	9.4g/dL	12 to 15 g/dL	c.92+5G>C	β^+ / β	Thalassaemia Minor
	HbA2	5.9%	1.5 to 3.5 %			
	MCV	72.1fL	80 to 96 fL			
	MCH	19.2pg	33 to 36 pg			
Father (37)	Hb	10.5g/dL	13 to 17 g/dL	c.79G>A p.Glu27Lys	HbE / β	HbE Trait
	MCV	70.8fL	80 to 96 fL			
	MCH	20.3pg	33 to 36 pg			
	HbE	71.40%	Absent			
Proband/ Son (5)	Hb	6.25g/dL	13 to 17 g/dL	c.79G>A p.Glu27Lys and 92+5G>C	HbE/β^+	HbE/ β – Thalassaemia*
	MCV	62.2fL	80 to 96 fL			
	MCH	20.1pg	33 to 36 pg			
	HbE	91.3%	Absent			
CVS		-	-	c.79G>A p.Glu27Lys (HbE)	HbE / β	HbE Trait

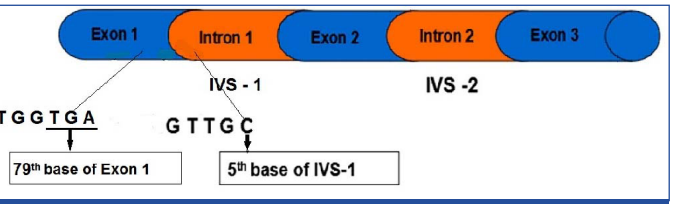
[Table/Fig-1]: Haematological indices of family (Rajasthani), MCV= Mean corpuscular volume, MCH = mean corpuscular haemoglobin, Hb= Haemoglobin, CVS= chorionic villus sampling.
Figures in Parenthesis indicate age in years. * Clinician suggested periodic transfusion.



[Table/Fig-2]: Showing DNA sequence analysis of family (Rajasthani); A. Mother with c.92+5G>C mutation (β+). B. Father with c.79 G>A P.Glu 27 lys (HbE trait), C. (Son) had (HbE, β+) co-inherited with/β-thalassaemia. D. CVS with HbE trait only. Nucleotide color, A= Green, T = Red, G= Black, C= Blue.



[Table/Fig-3]: Pedigree chart depicting autosomal recessive inheritance pattern of gene mutation transmission in a family.



[Table/Fig-4]: Showing Structure of HBB gene with mutation location.

as it had high HbE levels and altered normal Hb level (6.25 g/dl) along with red cell markers like MCV and MCH percent values as mentioned above. Clinician also suggested thalassaemia intermedia condition for periodic blood transfusion due to its probable severs condition. This kind of E/β compound heterozygosity is rare in Western India except one case in Gujarat [9] as for our knowledge is concerned in literature. But our case is the first to report from Rajasthan state reflecting its low incidence in this region too. Further, it is recommended that HbE/β thalassaemia patients do exhibit mild to severe anemic conditions variable from patient to patient availing periodic transfusions from childhood. It follows pedigree analysis showing autosomal recessive inheritance pattern [Table/Fig-3]. Molecular analysis indicated that both mutations of HbE and β thalassaemia in proband (son) are presented in Exon1 and Intron 1 of HBB gene whose segregation is slow probably for its low frequency [Table/Fig-4].

CONCLUSION

Co-inheritance of HbE-β thalassaemia case reported in a family of Rajasthan is rare in Western India. The proband (son five years) requires periodic blood transfusions as HbE level was higher with low Hb levels and changed phenotypic indices leading to thalassaemia intermedia condition. But parents and CVS possessed HbE and β thalassaemia traits only. Such families are suggested for prenatal screening for betterment in the society and to discourage marriages between such carriers.

ACKNOWLEDGEMENT

We thank all staff and clinicians of Supratech Micropath Laboratory, Ahmedabad for their continuous assistance in this work.

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PARTICULARS OF CONTRIBUTORS:
1. Chief Scientific Officer (CSO), Molecular Genomics, Supratech Micropath Laboratory and Research Institute, Ahmedabad, Gujarat, India.
2. Research Scientist, Supratech Micropath Laboratory and Research Institute, Ahmedabad, Gujarat, India.
3. Senior Scientist, Supratech Micropath Laboratory and Research Institute, Ahmedabad, Gujarat, India.
4. Laboratory Director, Supratech Micropath Laboratory and Research Institute, Ahmedabad, Gujarat, India.
5. Ex. Director, School of Sciences, Gujarat University, Ahmedabad, Gujarat, India.

NAME, ADDRESS, E-MAIL ID OF THE CORRESPONDING AUTHOR:
Dr. Sandhip C Shah,
Supratech Micropath Laboratory and Research Institute, Kedar Complex,
Opp. Krupa Petrol Pump, Near Parimal Garden, Ahmedabad-380006, Gujarat, India.
E-mail : supratech18@gmail.com

FINANCIAL OR OTHER COMPETING INTERESTS: None.

Date of Submission: **Dec 16, 2016**
Date of Peer Review: **Feb 14, 2017**
Date of Acceptance: **May 01, 2017**
Date of Publishing: **Sep 01, 2017**