Original Article

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Study of Mutations in β-Thalassemia Trait among Blood Donors in Eastern Uttar Pradesh

Internal Medicine Section

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ABSTRACT

Background: Knowledge on distribution of different mutations of thalassaemia, which are prevalent in a particular area, is a prerequisite for prenatal diagnosis.

Objectives: Studying mutations in β – thalassaemia trait among blood donors in eastern Uttar Pradesh, India.

Material and Methods: One thousand non–remunerated voluntary blood donors who were between 18 – 40 years of age, were included in the study. Both replacement and voluntary healthy blood donors were included. 4ml of venous blood was collected and it was stored at 4ºC. Complete Blood Count (CBC), Haemoglobinopathy Screening and Molecular Analysis by ARMS – PCR (Amplification Refractory Mutation System – PCR) were done. Screening for β thalassaemia was done in a blood bank by using D – 10, Bio Rad, which was based on High Performance Liquid Chromatography (HPLC).

Results: Twenty Eight subjects with β – thalassaemia trait were found among 1000 voluntary blood donors. IVS 1-5 (G-C) mutation was most common type (50%), followed by FS 8/9 (+G) 25% which was the second most common type. In our study, a rare mutation of CD 16 (-C) was also found. Out of 14 subjects who had IVS 1-5 (G-C) mutation (most common), six were from Varanasi (6/261) and five of them were Sindhis. It was seen that FS 41/42 (TCTT) mutation was distributed among all groups of populations which had higher prevalences of β-thalassaemia trait.

Conclusion: A comprehensive knowledge on beta thalassaemia mutations is necessary for determining a prenatal diagnosis. The occurrence of mutations may vary according to geographic region. Therefore, this study dealt with current problem of unknown mutations, in order to avoid complications.

Key words: Thalassaemia trait, Mutation, Haematology

INTRODUCTION

The cumulative gene frequency of haemoglobinopathies in India is 4.2%, with a population of over 1 billion [1]. Over 190 mutations have been recognized in thalassaemia, few being more common while others have rarely been encountered [2]. Moreover, certain mutations are more prevalent in a given population. No study has been done on the distribution of different mutations which are prevalent in eastern Uttar Pradesh and Bihar, especially in asymptomatic blood donors.

MATERIAL AND METHODS

The present study was carried out in the Department of Medicine, Department of Biochemistry and Blood Bank, Institute of Medical Sciences, Banaras Hindu University (BHU), Varanasi, India, during the period from May 2008 to May 2010. 1000 non – remunerated voluntary blood donors who were between 18-40 years of age, were included in the study. Blood Donor Selection criteria which was used was that which was prescribed in the AABB Technical Mannual [3]. Both replacement and voluntary healthy blood donors were included and complete physical examinations were done for all donors. 4 ml of venous blood was collected and it was stored at 4ºC. Complete Blood Count (CBC), Haemoglobinopathy Screening and Molecular Analysis by ARMS-PCR (Amplification Refractory Mutation System-PCR) were done. Screening for β thalassaemia was done in Blood Bank by using D-10, Bio Rad, which was based on High Performance Liquid Chromatography (HPLC).

STATISTICAL METHODS

Statistical analysis was done by using Fisher's exact probability test.

RESULTS

In our study, a majority of the individuals were between the age group of 21 and 30 years. Most common blood group was B^+, followed by the groups, O^+, A^+, B^-, A^-, AB^+,B^-, O^- and AB^-. 28 subjects with β-thalassaemia trait were found among 1000 voluntary blood donors of eastern part of UP; IVS 1-5 (G-C) mutation was most common type (50 %) , followed by FS 8/9(+G) 25 %, which was second most common type [Table/Fig-1]. In our study, a rare mutation, CD 16 (-C) was also found. Further, IVS 1-5(G-C) was found most commonly in Varanasi district [Table/Fig-2]. Out of 14 subjects who had IVS 1-5(G-C) mutation, six were from Varanasi and five of them were Sindhis. It was seen that FS 41/42 (-TCTT) mutation was distributed among all groups of populations which had higher prevalences of β-thalassaemia trait [Table/Fig-3].

DISCUSSION

Among the 1000 voluntary blood donors, 28 (2.8%) tested positive for β-thalassaemia trait and none tested positive for other haemoglobinopathies. A majority of study subjects were males (89.5%). The low number of female donors could be because of local social factors and physical health factors like anaemia, which may have barred them from donating blood. However, among those with the β-thalassaemia trait (n=28), 75% were males and 25% were

β-Globin mutations	β-Thalassemia traits (%)
IVS 1-5(G-C)	14 (50)
FS 8/9 (+G)	7 (25)
FS 41/42(-TCTT)	4 (14.3)
Del 619 bp	2 (7.1)
CD16(-C)	1 (3.6)

[Table/Fig-1]: Type of mutations among β-thalassemia trait (n=28)

District	Total No.	IVS 1-5 (G-C)	FS8/9 (+G)	Del 619 bp	FS41/42 (-TCTT)	Others
Varanasi	9	6	1	1	1	0
Ghazipur	3	2	0	0	1	0
Azamgarh	4	1	1	1	1	0
Jaunpur	2	0	2	0	0	0
Ballia	4	2	1	0	1	0
Bhadhoi	2	1	1	0	0	0
Deoria	2	1	0	0	0	CD-16C
Mau	2	1	1	0	0	0

[Table/Fig-2]: Distribution of mutations among β-Thalassemia trait in eastern Uttar Pradesh (n=28)

S.No.	Age	Sex*	Blood group	HbA_2 Level	Mutation
1.	20	F	A+	6.2	FS 41/42 (-TCTT)
2.	28	M	A-	5.9	FS 41/42 (-TCTT) Heterozygous
3.	21	M	B+	3.7	IVS 1–5 (G-C) Heterozygous
4.	34	M	AB-	7.3	CD16 (-C)
5.	19	F	A+	3.9	IVS 1-5 (G-C)
6.	27	M	B+	6.7	FS 8/9 (+G)
7.	23	F	O+	4.1	IVS 1-5 (G-C)
8.	20	M	A+	6.9	FS 8/9 (+G)
9.	30	M	O+	5.1	IVS 1-5 (G-C)
10.	24	F	B+	6.3	FS 41/42 (-TCTT)
11.	34	M	B+	3.8	Del 619 bp
12.	18	M	A+	7.2	IVS 1–5 (G-C)
13.	24	M	A+	6.1	FS 8/9 (+G)
14.	26	M	O+	8.4	IVS 1–5 (G -C)
15.	33	F	A+	4.3	IVS 1–5 (G-C)
16.	27	M	B+	8.1	F.S8/9 (+G)
17.	25	M	O+	4.5	Del 619 bp
18.	21	M	A+	6.2	FS 41/42 (-TCTT)
19.	28	M	O+	5.9	IVS 1–5 (G-C)
20.	38	F	B+	5.7	IVS 1–5 (G-C)
21.	23	M	O+	5.5	FS 8/9 (+G)
22.	36	M	A+	6.7	IVS 1–5 (G-C)
23.	21	M	B+	5.4	IVS 1–5 (G-C)
24.	26	M	O+	7.3	FS 8/9 (+G)
25.	19	M	A+	3.8	IVS 1-5 (G-C)
26.	35	F	O+	8.4	FS 8/9 (+G)
27.	30	M	A+	6.1	IVS 1-5 (G-C)
28.	24	M	B+	5.1	IVS 1-5 (G-C)

[Table/Fig-3]: Mutation detected among the blood donors having β-thalassemia trait (n=28)
*21 Males; 7 Females

females. A majority of blood donors who were under study were in the productive years (21-30 years) of their lives. However, since this study included only voluntary blood donors (they needed to be adults as per law), age distribution may not be a true representation of prevalence of thalassaemia in this population, but surely this study concluded about the importance of haemoglobinopathy among the so called healthy blood donors. The prevalence of different mutations varies significantly in different regions of India [4-9]. The IVS 1-5 mutation was the commonest mutation which was found in the Indian populations and its prevalence varied from 22.8 to 81.4% in different regions of India [10]. In our study, 28 subjects with the β-thalassaemia trait were found among 1000 voluntary blood donors of eastern part of UP; IVS 1-5 (G-C) mutation was most common type (50%) [Table/Fig-1], which was similar to that which was found in previous studies, followed by FS 8/9(+G) 25%, which was second most common type, which was not found in previous studies [4,5,11,12]. This may be due to the selective population of voluntary blood donors who were between age 18 and 40 years of age and it does not reflect the actual population. However, the latter is one of the five common types of mutations which are found in India [4,11]. In our study and in a previous study [4] also, it was seen that FS 41/42 (-TCTT) mutation was distributed among all groups of populations, which had a higher prevalence of β-thalassaemia trait. In our study, a rare mutation, CD 16 (-C) was also found. In the study of Vaz et al, the latter was found predominantly in Brahmins, as compared to that in Rajputs and Vaishyas (mainly in Gujarat) [12]. Further, in our study, IVS 1-5 (G-C) was found most commonly in Varanasi district [Table/Fig-2]. Out of 14 subjects who had IVS 1-5 (G-C) mutation, six were from Varanasi and five of them were Sindhis. The latter were not the natives of Varanasi but they had migrated from outside. In an earlier study from India, it was seen that the mutation, IVS 1-5(G-C) was most common among Sindhis [13], while in other studies, 619 bp deletion mutation was found to be the commonest β-thalassaemia mutation which was observed in patients who originated from Sindh, Gujarat or from among the families which had migrated from Pakistan [11,14].

CONCLUSION

Thalassaemia is a syndrome of inherited haemoglobin disorders, which is characterised by a quantitative deficiency of functional beta and alpha-globin chains. Routine investigative modalities for identification of β-thalassaemia include low red cell values (MCV, MCH, RDW) on a complete blood picture (CP), an altered erythrocyte morphology and increased Hb A2 levels on high performance liquid chromatography (HPLC) or Hb electrophoresis. This study was conducted to check the spectrum of β-thalassaemia mutations and their association with individuals in Uttar Pradesh, India.

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