A CROSS-SECTIONAL GENOTYPIC STUDY OF THALASSEMIA CHILDREN ENROLLED IN THALASSEMIA CLINIC IN A TERTIARY CARE HOSPITAL, MYSURU

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KEYWORDS

Thalassemia; Genetic mutations; Genotype; Phenotype; India

ABSTRACT

Background: Thalassemia is a genetic disorder affecting hemoglobin production, with a high prevalence in India. Approximately 240 million people worldwide are carriers of β-thalassemia, with 30-40 million carriers in India. **Objectives:** To identify the different types of genetic mutations present in children with thalassemia and to correlate genotype with phenotype. **Methods:** A cross-sectional descriptive study was conducted among 50 patients enrolled in the thalassemia clinic at a tertiary care hospital in Mysuru. A blood sample was collected from all the children enrolled in thalassemia clinic of JSS Hospital, Mysuru coming for routine blood transfusion. The sample was sent to the laboratory for genetic analysis. **Results:** The majority of patients were older children and adolescents, with 48% being over 10 years old. The IVS-I-5 (G>C) mutation was the most common, found in 54% of patients. The most common variant, C.92+5G>C, found in 72% of the patients, suggests a significant concentration of this mutation in the studied population. Other variants, like C.47G>A (12%) and C*110T>C (6%), are less frequent, indicating that while the C.92+5G>C mutation is predominant, there is still notable genetic heterogeneity The most prevalent genotype was B+/B+, found in 62% of patients. **Conclusion:** In conclusion, this study reveals a significant concentration of the IVS-I-5 (G>C) mutation and the C.92+5G>C variant among the studied population, highlighting the importance of genetic testing in diagnosis and treatment planning. The correlation between genotype and phenotype

was also established, with the B+/B+ genotype being the most prevalent and associated with a less severe form of thalassemia. The study underscores the significance of genetic inheritance patterns in the manifestation of the condition and emphasizes the need for personalized treatment strategies based on individual genetic profiles.

Introduction

Thalassemia is a crippling, chronic illness that affects around 200 million people worldwide.[1]Hereditary mutations in the gene that produces the globin chains—essential building blocks of healthy adult hemoglobin—cause the illness. These mutations cause erythropoiesis to be inefficient, which in turn causes anaemia.[2]

Approximately 240 million people worldwide are carriers of β- thalassemia.[3] Thirty to forty million people in India are carriers of the disease, and approximately 12,000 babies born there each year have a serious form of the illness[4,5] making up 10% of all babies worldwide.[6] The most frequent inherited haemolytic anaemias worldwide are Thalassemia and hemoglobinopathies, which are particularly widespread in Southeast Asian nations.

Mutations in the globin genes induce Thalassemia, which is characterized by an imbalance in globin production and inefficient erythropoiesis.[7] In the Asian population, homozygous beta-thalassemia, beta- thalassemia/Haemoglobin (Hb) E disease, and Hb H disease are the three main forms of Thalassemia illnesses. In the population, carriers of alpha and beta Thalassemia are found at frequencies of roughly 20–30% and 3–9%, respectively. The frequency of Hb E, a prevalent Hb variation in the area, ranges from 10% to 53%. Furthermore, it is thought that between 1% and 8% of people have Hb Constant Spring, a prevalent non-deletional alpha- thalassemia.[8]

The frequency of β-thalassemia mutations in the Indian population is not well-documented. As a result, our study assisted us in determining the frequent mutations found in this Thalassemia belt of India as well as whether any distinct mutations are more common in Mysuru than in the Indian population.

Objectives

To know the different types of genotypes in thalassemia and to compare genotype with phenotype of thalassemia children enrolled at thalassemia clinic in a tertiary care hospital, Mysuru.

Materials and method

The present cross-sectional descriptive study was conducted among 50 patients who were enrolled in thalassemia clinic in tertiary care referral hospital, Mysuru over a period of one year from September 2022 to September 2023. The study utilized convenience sampling, where all patients enrolled in the thalassemia clinic at a tertiary care hospital in Mysuru during the study period were included in the sample.

The study included all children enrolled in the thalassemia clinic at a tertiary care referral hospital in Mysuru during the study period. Children with a combination of thalassemia and any other haemoglobinopathy were excluded from the study.

Study assessments of end points

The study seeks to identify the different types of genetic mutations present in children enrolled in the thalassemia. Additionally, the study also explored the correlation between the genotype and phenotype in thalassemia children, examining how specific genetic mutations influence the severity and characteristics of the disease.

A blood sample was collected from all the children enrolled in thalassemia clinic of JSS Hospital, Mysuru coming for routine blood transfusion. The sample was sent to the laboratory for genetic analysis.

Data Analysis: All collected data were systematically entered into an Excel spreadsheet to ensure organization and accuracy. For detailed statistical analysis, the data were then imported into SPSS version 20. Descriptive statistics was done to summarize the data, and inferential statistics to identify patterns, correlations, and significant differences. This

approach allowed for a comprehensive evaluation of the study's objectives, ensuring robust and reliable results.

Results

Table 1: Distribution depending on age group

Age in years	Frequency	Percentage
<1	4	8
1 to 10	22	44
>10	24	48
Total	50	100

Table 2: Distribution depending on genetic mutation

Genetic mutation	Frequency	Percentage
IVS-I-5 (G>C)	27	54
CODON 15(G>A), IVS-I-5(G>C)	5	10
POLY A(T->C)	3	6
CODON 15(G>A), IVS-I-6(G>C)	1	2
88 C>T	1	2
619 BP DELETION	1	2
CAP +1 (A>) SILENT	1	2
HBE, IVS-I-5G>C	1	2
IVS-II-837(T->G)	1	2
No pathogenic mutation	1	2

The age distribution of the patients shows (table 1) that the majority are older children and adolescents, with 48% being over 10 years old. The IVS-I-5 (G>C) mutation, found in 54% of the patients, is a common mutation associated with certain types of thalassemia (table 2). The presence of other mutations like CODON 15(G>A), IVS-I-5(G>C) in 10% and POLY A(T->C) in 6% indicates genetic diversity in the patient population. These mutations can affect the severity and clinical presentation of the disease, influencing treatment decisions and prognosis. The data suggest that while some mutations are prevalent, there is also a variety of less common genetic variants in this population.

Table 3: Distribution depending on HGVS (Human Genome Variation Society)

HGVS	Frequency	Percentage
C.92+5G>C	36	72
C.47G>A	6	12
C*110T>C	3	6
C.-138 C>T	1	2
C.20 A>T	1	2
C.316-14T>G	1	2

The HGVS (Human Genome Variation Society) nomenclature provides a standardized way to describe genetic mutations (table 3). The most common variant, C.92+5G>C, found in 72% of the patients, suggests a significant concentration of this mutation in the studied population. Other variants, like C.47G>A (12%) and C*110T>C (6%), are less

frequent, indicating that while the C.92+5G>C mutation is predominant, there is still notable genetic heterogeneity. Understanding these mutations is essential for accurate diagnosis and personalized treatment strategies.

Table 4: Distribution depending on genotype

Genotype	Frequency	Percentage
B+/B+	31	62
B+/B0	12	24
B0/B0	6	12
HBS/HBS	1	2
Total	50	100

The most prevalent genotype, B+/B+, found in 62% of the patients, typically indicates a less severe form of thalassemia compared to B0/B0, which is associated with more severe disease (table 4). The presence of 24% of B+/B0 and B0/B0 genotypes in 12% of patients suggests a mix of severity within the population. The diversity in genotypes highlights the importance of genetic testing in determining the course of the disease and tailoring treatment accordingly. The rare HBS/HBS genotype (2%) indicates the presence of sickle cell disease in a small subset of patients, further adding to the complexity of the genetic landscape.

Table 5: Distribution depending on allele status

Allele	Frequency	Percentage
Homozygous	37	74
Heterozygous	13	26
Total	50	100

The predominance of homozygous alleles (74%) suggests that most patients inherited the same mutation from both parents, leading to a more consistent and often severe clinical presentation (table 5). In contrast, the heterozygous status in 26% of the patients could indicate a milder form of the condition or a carrier state, depending on the specific mutations involved. This distribution underscores the significance of genetic inheritance patterns in the manifestation of the condition.

Table 6: Distribution depending on clinical significance

Diagnosis	Frequency	Percentage
T inter/Major	48	96
Sickle cell disease	1	2
Normal	1	2
Total	50	100

The most common clinical diagnosis was Thalassemia Intermedia/Major, affecting 96% of the patients (table 6). This indicates a high burden of severe thalassemia, which requires ongoing medical management. The presence of sickle cell disease in 2% of patients suggests that while it is less common, it still contributes to the overall disease burden.

Table 7: Association between genotype and various phenotypic parameters

Genotype	B+/B+ (n=31)	B+/B0 (n=12)	B0/B0 (n=6)	HBS/HBS (n=1)	P value
Mean age in years	10.6	10.7	8.8	18	0.5
Male	19	8	5	1	0.65
Female	12	4	1	0	
Consanguineous marriage	23	8	5	1	0.8
Mean height in cms	100.1	110.1	97.5	89	0.45
Mean weight in kgs	17.2	21.7	17.5	18	0.21
Mean liver size	3.7	3.7	3.8	4	0.2
Mean spleen size	7.6	7.5	7.6	10.4	0.4
Transfusion frequency	17	16.7	14.1	12	0.34
Family history present	16	7	5	0	0.33
Homozygous	31	0	5	1	<0.0001*
Heterozygous	0	12	1	0	

Table 7 shows association between genotype and various phenotypic parameters. In the study, the mean age of patients varied slightly by genotype, with B+/B+ at 10.6 years, B+/B0 at 10.7 years, B0/B0 at 8.8 years, and HBS/HBS at 18 years. Gender distribution showed a predominance of males in all genotypes except HBS/HBS, which had one male and no females. Consanguineous marriages were reported for 23 patients with B+/B+, 8 with B+/B0, 5 with B0/B0, and 1 with HBS/HBS. The average height was highest in B+/B0 patients (110.1 cm) and lowest in HBS/HBS patients (89 cm), while mean weight and liver size were relatively similar across genotypes, except for slightly higher values in B+/B0 and HBS/HBS groups. Mean spleen size was also largest in HBS/HBS patients (10.4 cm). Transfusion frequency was highest in B+/B0 patients (16.7 times) and lowest in HBS/HBS patients (12 times). Family history of thalassemia was present in 16 patients with B+/B+, 7 with B+/B0, 5 with B0/B0, and none with HBS/HBS. Genotypically, all B+/B+ patients were homozygous, B+/B0 patients were exclusively heterozygous, and a few B0/B0 and HBS/HBS patients were homozygous. A statistically significant P value is less than 0.05.

Discussion

The genetic analysis shows that the IVS-I-5 (G>C) mutation is the most common among the patients, found in 54% of cases. The presence of other mutations, such as CODON 15(G>A), IVS-I-5(G>C) in 10% and POLY A(T->C) in 6%, reflects the genetic diversity within the population. These mutations can influence the severity and clinical presentation of the disease, impacting treatment decisions and prognosis. The data suggests that while certain mutations are prevalent, there is also a notable presence of less common genetic variants that contribute to the condition's complexity. The HGVS (Human Genome Variation Society) nomenclature findings, particularly the high prevalence of the C.92+5G>C variant (72%), highlight a significant concentration of this mutation in the study population. Other variants, such as C.47G>A (12%) and C*110T>C (6%), are less common, indicating genetic heterogeneity. Understanding these mutations is crucial for accurate diagnosis and personalized treatment strategies, as different mutations may respond differently to treatment or have varying prognostic implications.

Study by Tamer Hassan et al[9] showed that the most prevalent mutations were IVS 1–1, IVS 1–110, and IVS 1–6, occurring in 26.7%, 22.6%, and 18.5% of cases, respectively. Other notable mutations include IVS 11–745 at 9.6%, Codon 39 at 8.9%, and Codon 5 at 3.4%. Less common mutations include Promoter 87, Codon 15, IVS 11–848, Codon 37, Codon 44, and Codon 27, with frequencies ranging from 0.7% to 2.7%. While IVS 1–1[G > A], IVS 1– 110[G > A], and IVS 1–6[T > C] are the most commonly reported mutations in various Egyptian studies, their relative frequencies differ across studies. For instance, one study may report IVS 1–1 as the most frequent mutation, followed by IVS 1–110 and IVS 1–6, while another might rank IVS 1–110 as the most prevalent, with IVS 1–1 and IVS 1–6 in different positions. These variations in mutation frequency rankings can be attributed to differences in sample populations, geographic regions, or methodologies used in the studies.[10-12]

Study by Meloni A et al[13] showed that total of thirty-three distinct genotypes were found; homozygous CD39, CD39/IVS-1,110, and homozygous IVS-1,110 were the most prevalent. In Mediterranean populations, Huisman et al[14] identified IVS 1–110[G > A], IVS 1–6[T > C], and IVS 1–1[G > A] as the most common mutations, along with promotor 87[C > G], IVS 11–745[C > G], and C39[C > T]. Conversely, in the Middle East, the most prevalent mutations were C8[-AA], C8, /C9[+G], IVS 1–5[G > C], C39[C > T], C44[-C], and IVS 11–828[C > A]. This variation highlights regional differences in mutation frequency within these broader geographic areas. Study by Panigrahi I et al[15]did an interstate comparison in India and found that mutations identified across various regions of India. Among these, the IVS-I-5 (G→C) mutation is the most prevalent beta-thalassemia allele in the Indian population. The 619 bp deletion mutation is notably prevalent among Sindhis and Lohanas, particularly from Gujarat. In a study by Vaz et al[16] of 1233 carriers, rare beta-thalassemia mutations were identified in 87 individuals (7.06%). Additionally, a high occurrence of the IVS-1-1 (G→T) mutation was observed among Sindhis (25.5%), Punjabi Hindus (34.7%), and Lohanas (31.2%).

The genotype distribution, with B+/B+ being the most common (62%), suggests that a majority of patients have a less severe form of the condition compared to those with B0/B0 genotypes, which are associated with more severe disease. The presence of 24% of B+/B0 and B0/B0 genotypes in 12% of patients indicates a mix of severity levels within the population. This diversity in genotypes underscores the need for genetic testing to guide treatment decisions and predict disease progression. Study by Tamer Hassan et al[101] showed that most common genotype among the patients was β+β+, found in 49.3% of cases, followed by β°β°, which occurred in 37.0% of patients. The β°β+ genotype was less frequent, observed in 13.7% of the cases. Study by Meloni A et al[13] showed that genotypes were classified based on their phenotypic expression as β+ or β0, and patients were grouped accordingly. The categories were: homozygous β+ with 19 patients (27.9%), compound heterozygous β0β+ with 24 patients (35.3%), and homozygous β0 with 25 patients (36.8%). Al Akhras et al[10] observed that patients with the β°β° genotype exhibited a more severe phenotype. A recent study by Garewal et al [17] revealed a significantly high prevalence (46%: 41 out of 88) of the mild β++ promoter mutation -88 (C→T) among Punjabis.

The predominance of homozygous alleles (74%) among the patients suggests a significant genetic influence, as these patients inherited the same mutation from both parents, leading to a more severe clinical presentation. The presence of heterozygous alleles in 26% of the patients may indicate a milder form of the condition or a carrier state, depending on the specific mutations involved. This distribution highlights the importance of understanding inheritance patterns and their impact on the disease's manifestation and severity. Study by Tamer Hassan et al[9] showed that homozygous IVS 1–1 was the most prevalent, occurring in 19.17% of the patients. This was followed by homozygous IVS 1–110, found in 15.06% of cases, and homozygous IVS 1–6, present in 10.95% of the patients. These genotypes represent the most common genetic variations in the cohort, indicating their significant role in the population studied. Elmezayen et al[12]

reported that 51% of patients had homozygous mutations, while 28% had compound heterozygous mutations. The most prevalent genotypes identified were homozygous IVS 1–6[T > C], IVS 1–110[G > A], and promotor 87[C > G], as well as compound genotypes IVS 1– 6[T > C]/IVS 1–110[G > A] and IVS 1–6[T > C]/IVS 11–848[C > A], occurring in 15%, 13%, 6%, 6%, and 6% of patients, respectively. In contrast, El-Shanshory et al[18] found that compound heterozygous mutations were more prevalent than homozygous mutations among their patients.

The diagnosis distribution reveals that an overwhelming majority of the patients (96%) are diagnosed with Thalassemia intermedia or major. This high percentage underscores the prevalence of this severe form of the disease in the study population. The small percentage of patients diagnosed with Sickle cell disease (2%) and those categorized as normal (2%) suggests that while Thalassemia is the primary condition being studied, other Haemoglobinopathies like Sickle cell disease is present but less common. This distribution emphasizes the need for targeted interventions and management strategies focused on Thalassemia, given its dominance in the patient population. Study by Tamer Hassan et al[9] showed that Transfusion-dependent Thalassemia, also known as Thalassemia major, affects 83.6% of patients and calls for frequent blood transfusions. 16.4% of the remaining individuals had Thalassemia intermedia, which is not dependent on transfusions, which does not require frequent transfusions but may still need occasional treatment. This shows that thalassemia major is more common in this group. Winichagoon et al[19] categorized the patients into three groups based on disease severity: 47 patients classified as light, 55 as intermediate, and 43 as severe. Camaschella et al. [20] examined a cohort of 292 Italian patients, of whom 127 had Thalassemia major and 165 had Thalassemia intermedia as their diagnosis. This research provided insights into the clinical and genetic characteristics of both forms of the disease within this population, allowing for a detailed comparison of the two types of thalassemia.

The study revealed that the mean age of patients was similar across B+/B+ (10.6 years) and B+/B0 (10.7 years), slightly lower for B0/B0 (8.8 years), and significantly higher for

HBS/HBS (18 years). Gender distribution showed a predominance of males in most genotypes, with B+/B+ and B+/B0 having 19 and 8 males, respectively, while HBS/HBS had only one male. Consanguineous marriages were most common in the B+/B+ group (23 cases). Average height was highest in B+/B0 patients (110.1 cm) and lowest in HBS/HBS patients (89 cm), with similar mean weights and liver sizes across groups. Spleen size was largest in HBS/HBS patients (10.4 cm). Transfusion frequency was highest in B+/B0 patients (16.7 times) and lowest in HBS/HBS patients (12 times). Family history of thalassemia was noted most frequently in the B+/B+ group. Study by Tamer Hassan et al[9] showed that compared to individuals with the β°β+ and β+β+ genotypes, those with the β°β° genotype showed a greater prevalence of hepatomegaly and hepatitis C (81.5% and 40.7% vs. 36.1% and 11.1%, respectively). They also had increased rates of cardiac complications, growth retardation, hypogonadism, and hypothyroidism, as well as a higher incidence of splenectomy (40.7% vs. 2.8% in β+β+). Additionally, β°β° patients showed significantly higher liver iron content (27.3 mg/g dw) and lower cardiac T2* values (21.3 ms), indicating greater iron overload compared to the β°β+ and β+β+ genotypes. Shamoon et al[21] identified IVS-I-6 (T > C) as common mutation, occurring in 34.6% of patients with thalassemia intermedia.

This study has few limitations. Firstly, the small sample size and limited geographical scope may restrict the generalizability of the findings, making it challenging to apply the results to broader Thalassemia populations. Secondly, the cross-sectional design provides only a glimpse of genotypic and phenotypic characteristics at a single point in time, limiting the ability to assess disease progression, long-term outcomes, or causative relationships.

Conclusion

In conclusion, this cross-sectional study provides valuable insights into the genetic characteristics and clinical presentation of thalassemia children enrolled in a tertiary care hospital in Mysuru. The study reveals a significant concentration of the IVS-I-5 (G>C) mutation and the C.92+5G>C variant, highlighting the importance of genetic testing in

diagnosis and treatment planning. The correlation between genotype and phenotype was also established, with the B+/B+ genotype being the most prevalent and associated with a less severe form of thalassemia. The study underscores the significance of genetic inheritance patterns in the manifestation of the condition and emphasizes the need for personalized treatment strategies based on individual genetic profiles.

References

1. Biswas B, Naskar NN, Basu R, Dasgupta A, Paul B, Basu K. Knowledge of the caregivers of thalassemic children regarding thalassemia: A cross-sectional study in a tertiary care health facility of eastern India. Iraqi J Hematol 2018;7:49-54.
2. Kasper D, Fauci A, Hauser S, Longo D, Jameson J, Loscalzo J, editors. Harrison's Principles of Internal Medicine. Vol. 19e. New York: McGraw-Hill, Medical Publication Division; 2015. p. 633-6.
3. Cao A, Kan YW. The prevention of thalassemia. Cold Spring Harb Perspect Med 2013;3:a011775.
4. World Health Organisation. Management of Haemoglobin Disorders: Report of a Joint WHO-TIF Meeting, Nicosia, Cyprus, 16-18 November, 2007; 2008. Available from: http://www.apps.who.int/iris/bitstream/10665/43969/1/9789241597128_en g.pdf.
5. Mohanty D, Colah RB, Gorakshakar AC, Patel RZ, Master DC, Mahanta J, et al. Prevalence of β-thalassemia and other haemoglobinopathies in six cities in India: A multicentre study. J Community Genet 2013;4:33-42.
6. Varawalla NY, Old JM, Sarkar R, Venkatesan R, Weatherall DJ. The spectrum of beta-Thalassemia mutations on the Indian subcontinent: The basis for prenatal diagnosis. Br J Haematol 1991;78:242-7.
7. Charoenkwan P, Komvilaisak P, Thepsuthummarat K, Seksarn P, Torcharus K. Causes of Hospitalizations in Pediatric Patients with Thalassemia under the National Health Coverage Scheme in Thailand. Thalassemia Reports 2024;14:10-17.
8. Fucharoen S, Weatherall DJ. Progress Toward the Control and Management of the Thalassemias. Hematol Oncol Clin North Am 2016;30:359-371.
9. Tamer Hassan, Mohamed Badr, Usama El Safy, Mervat Hesham, Laila

Sherief and Marwa Zakaria. β-Thalassemia: Genotypes and Phenotypes. Epidemiology of Communicable and Non-Communicable Diseases - Attributes of Lifestyle and Nature on Humankind. 2016.

10. Al-Akhras A, et al. Impact of genotype on endocrinal complications in β-thalassemia patients. Biomed Rep. 2016;4:728–736.
11. Soliman OE, et al. Reverse hybridization Strip Assay detection of beta-thalassemia mutations in northeast Egypt. Hematology. 2010;15:182–
12. Elmezayen AD, Kotb SM, Sadek NA, Abdalla EM. β-Globin Mutations in Egyptian Patients With β-Thalassemia. Lab Med. 2015;46:8–13.
13. Meloni A, Pistoia L, Ricchi P, Putti MC, Gamberini MR, Cuccia L, Messina G, Massei F, Facchini E, Righi R, et al. Link between Genotype and Multi-Organ Iron and Complications in Children with Transfusion-Dependent Thalassemia. Journal of Personalized Medicine. 2022; 12(3):400.)
14. Huisman, T.H.J., Carver, M.F.H. & Baysal E. In Beta-thalassemia; nondeletional mutants. (Eds Huisman, T. H. J., Carver, M. F. H., Baysal, E., Marianne, F. H.). A syllabus of thalassemia mutations. Augusta, GA: The Sickle Cell Anaemia Foundation. 1–309 (1997).
15. Panigrahi I, Marwaha RK. Mutational spectrum of thalassemias in India. Indian J Hum Genet. 2007 Jan;13(1):36-7.
16. Vaz FE, Thakur (Mahadik) CB, Banerjee MK, Gangal SG. Distribution of β-thalassemia mutations in the Indian population referred to a diagnostic center. Hemoglobin. 2000;24:181–94.
17. Garewal G, Das R, Ahluwalia J, Marwaha RK, Varma S. Nucleotide -88 (C-T) promoter mutation is a common beta-thalassemia mutation in the Jat Sikhs of Punjab, India. Am J Hematol. 2005;79:252–6.
18. El-Shanshory M, et al. Spectrum of Beta Globin Gene Mutations in Egyptian Children with β-Thalassemia. Mediterr J Hematol Infect Dis. 2014;6:e2014071.

19. Winichagoon P, Fucharoen S, Chen P, Wasi P. Genetic factors affecting clinical severity in beta-thalassemia syndromes. J Pediatr Hematol Oncol. 2000;22:573–580

20. Camaschella C, et al. Genetic interactions in thalassemia intermedia: analysis of beta-mutations, alpha-genotype, gamma-promoters, and beta- LCR hypersensitive sites 2 and 4 in Italian patients. Am J Hematol. 1995;48:82–87.

21. Shamoon RP, et al. Molecular Basis of β-Thalassemia Intermedia in Erbil Province of Iraqi Kurdistan. Haemoglobin. 2015;39:178–183.