Original Article

The Prime Mutation of β-Thalassemia and Disease Severity in (Wasit and Maysan) South of Iraq

Anwer AL-SARRAY[1]*, Hedef El-YASSIN[2], ALI JAWAD[3]

[1]Department of Pharmacy, Alkut collage university, Iraq.
Email: anwarhilal2015@gmail.com
[2]Department of clinical Biochemistry, Collage of Medicine Baghdad university, Iraq.
Email: hedefelyassin@gmail.com
[3]Department of Hematology, Collage of Medicine Baghdad university, Iraq.
Email: almothafara@gmail.com

Abstract

Thalassemia is a hereditary disease caused by a mutation in the globin gene, resulting in anemia as a risk factor for patients' lives. There are different types of mutations in the globin gene. Despite the types of mutations differing from area to area due to migration and traditions between worlds, there is a need to recognize these different mutations in order to control the disease, and to identify the spectrum of the most common mutations and the relationship between these mutations and the severity of the disease.

Keywords: Iraq, Thalassemia, Globin, Mutation.

Introduction

Thalassemia is a variety of blood disorders in which hemoglobin genes are disrupted, resulting in inefficient erythropoiesis. Genetic disorders caused by decreased production of the alpha or beta chains of hemoglobin.[1,2]

Hemoglobin is composed of two proteins, alpha and beta. When the body fails to produce either of these proteins in sufficient quantities, anemia develops in early childhood and persists throughout life. However, thalassemia is inherited in an autosomal recessive manner.[3,4] Beta-thalassemia is caused by mutations or, in rare cases, deletions of the beta-globin gene (HbB) on chromosome.[5] These are usually point mutations.[3,6] Beta (+) for decreased production and beta (0) for absent production is the genotypic variability of beta-globin synthesis.[7,8] The most common mutations found in northern Iraq were IVS-II -1 (G > A), IVS-I-6 (T > C), IVS-I-I (G > A), codon 8 (AA), codon 8/9 (+G), IVS-I-110 (G > A), and codon 5 (CT), which accounted for approximately 82% of all described alterations. Codon 39 (C > T), codon 44 (C), 14 IVS-I-5 (G > C), codon 82/83 (G), and IVS-I-128 (T > G) are among the rarer alterations.[9] The 0-thalassemia is typically caused by substantial deletions that effect both the HBA2 and HBA1 genes; however,(+)-thalassemia can be caused by either an extra mutation that affects only one HBA gene or a point mutation that decreases -globin chain production.[10] Homozygosity for the β-globin gene IVS-1-110[G>A] c.93-21G>A and codone 27[G>T] Knossos c.82G>T allele was the major contributing factor for the beta thalassemia in the study subjects so ots mor sever motility and morbidity . The role of IVS 1.1 [G>A] c.92+1G>A and IVS 1.6 [T>C] c.92+6T>C mutations in of the beta thalassemia was observed in Heterozygous mutant. The IVS 1.5 [G>C] c.92+5G>C mutation was diagnosed in one patient in and hetrozygot In heterozygous individuals, -globin gene deletions, particularly those in promoter regions, are frequently associated with high levels of hemoglobin A2 (HbA2). Furthermore, fetal hemoglobin (HbF) levels are elevated due to -globin gene deletions. However, significant deletions of the - and -globin genes have been seen in certain carriers with normal HbF levels. Because most deletions may be missed by DNA sequencing, identifying deletions in the -globin gene by alternative techniques is critical.[11]

Access this article online

Quick Response Code:

Website: www.jnsbm.org

DOI: https://doi.org/10.4103/jnsbm.JNSBM_14_2_18

Address for Correspondence: Department of Pharmacy, Alkut collage university, Iraq
Email: anwarhilal2015@gmail.com

Submitted: 25th September, 2023 **Received:** 05th October, 2023
Accepted: 24th October, 2023 **Published:** 12th November, 2023

How to cite this article: AL-SARRAY A, El-YASSIN H, JAWAD A. The Prime Mutation of β-Thalassemia and Disease Severity in (Wasit and Maysan) South of Iraq. J Nat Sc Biol Med 2023;14:221-224

Several studies have been conducted in Iraq to identify the types of mutations that cause thalassemia based on their geographic distribution in different parts of the country. IVS1nt-5 mutation (3.61%), codon-15 mutation (3.61%), -88 mutation (2.4%), and codon-30 mutation (2.4%) were found in 2 (2.4%) of patients, while codon-8/9 mutation was found in 5 (6.1%) of patients. Heterozygosity for codon-15 was found in 28 (33.7%) of patients, while IVS1nt-5 and codon 8/9 mutations were found in 15 (18%) and 9 (10.8%) of patients, respectively. In addition, heterozygosity for the -88 mutation and codon 30 mutation was found in 8 (9.6%) and 6 (7.2%) of patients, respectively. Only 2 (2.4%) of patients in southern Iraq have codon 41/42.[3] Purpose of the study to detect the common mutation in South of Iraq.

Method

Three milliliters of whole blood were drawn from each participant to analyze the common mutation using (Vienna lab strip assay kits).

Participants: The study included 100 beta-thalassemia patients from thalassemia centers in Wasit and Maysan.

Data Collection and Analysis

The procedure consists of three steps:

1. DNA extraction.
2. Amplification by PCR.
3. The amplification products are hybridized with a test strip consisting of allele-specific oligonucleotide probes arranged as an array of parallel lines. The test detects 22 -globin mutations: - 101 [C>T], - 87 [C>G], - 30 [T>A], codon 5 [-CT], codon 6 [G>A] HbC, codon 6 [A>T] HbS, codon 6 [-A], codon 8 [-AA], codon 8/9 [+G], codon 15 [TGG>TGA], codon 27 [G>T] Knossos, IVS 1.1 [G>A], IVS 1.5 [G>C], IVS 1.6 [T>C], IVS 1.110 [G>A], IVS 1.116 [T>G], IVS 1.130 [G>C], codon 39 [C>T], codon 44 [-C], IVS 2.1 [G>A], IVS 2.745 [C>G], IVS 2.848 [C>A].

The Scale

Findings

Table 1. The results of Beta Globin Strip Assay analysis

Characteristic	Normal n (%)	Heterozygous mutant n (%)	Homozygous mutant n (%)
codon 27 [G>T] Knossos c.82G>T	(6%)	(0%)	(94%)
IVS 1.1 [G>A] c.92+1G>A	(0%)	(94%)	(6%)
IVS 1.5 [G>C] c.92+5G>C	(34%)	(66%)	(0%)
IVS 1.6 [T>C] c.92+6T>C	(0%)	(100%)	(0%)
IVS 1.110 [G>A] c.93-21G>A	(6%)	(0%)	(94%)

Table 2. the genotype and disease severity

Mutation	Genotype	Disease severity
codon 27 [G>T] Knossos c.82G>T	Homozygous mutant n (%)	Sever
IVS 1.1 [G>A] c.92+1G>A	Heterozygous mutant n (%)	sever
IVS 1.6 [T>C] c.92+6T>C	Heterozygous mutant	Sever
IVS 1.5 [G>C] c.92+5G>C	Heterozygote	mild
IVS 1.110 [G>A] c.93-21G>A	Homozygous mutant	sever

Figure 1: Beta Globin Strip Assay sheet analysis.

Discussions

In the present study, a total of five of the most common mutations identified by strip assay kits (ViennaLab-Austria) were detected (IVS 1.6 [T > C] c.92+6T > C, IVS 1.1 [G > A] c.92+1G > A, IVS 1.5 [G > C] c.92+5G > C) were present in the heterozygous genotype of beta-thalassemia, whereas (IVS 1.110 [G > A] c.93-21G > A, codon 27 [G > T] Knossos c.82G > T) were present in the homozygous genotype.

Eissa *et al.*[5] noted that the IVS 1.6 [T > C] c.92+6T > C mutation is one of the most common in northern Iraq in patients with thalassemia. It is common in all Arab Mediterranean countries.[12]

IVS 1.5 [G > C] c.92+5G > C was reported in 66 cases; these results are virtually identical to those reported by Al-Badran *et al.*[3]; it is the most common mutation in Basra, southern Iraq. The IVS 1.5 [G > C] c.92+5G > C mutation was the most common in the United Arab Emirates and was found at very high rates in the Indian subcontinent and in people bordering India.[3]

IVS 1.1 [G > A] The c.92+1G > A mutation found in this study was the same as that found in Rezaee *et al.*[13]. IVS-I-1 (G > A) was the most common of the -globin mutations found in Kurdistan and southwestern Iran near the Arabian Peninsula. In the current study, IVS-I-110 (G/A) was found to homogeneously genotype the beta-globin mutation. Similar to the results of Eissa *et al.*[5], this is the most common mutation in northern Iraq (Mosul), as well as the most common mutation in neighboring Turkey, Syria, Lebanon, Jordan and Upper Egypt.[14] According to recent studies, it is more common in central Iraq.[15]

The mutation in beta-globin in homogeneous patients was codon 27 [G > T] Knossos c.82G > T (Hb Knossos), which triggers a cryptic splice site in the -globin gene that competes with the normal splice site, resulting in decreased Hb synthesis. It has been described in association with several -thal mutations such as IVS-I-1 (G > A) and IVS-I-110 (G > A).[16] Hb Knossos has been identified in Egypt, Tunisia, Jordan and Gaza.[16] The prevalence is high in all Arab Mediterranean countries, but lower in the Gulf countries.[12] The prevalence of codon 27, G > T (Hb Knossos) was 1.40 in Egypt and 0.10 in Turkey.[1] These studies differ from the current study in that codon 27, G > T (Hb Knossos) is more prevalent at the current study site. Iraq has a multiethnic population that has been affected by numerous invasions and migrations throughout history. Iraq's strategic location as a crossroads between Western and Eastern civilizations, as well as the high degree of consanguinity among the Iraqi population, has led to Iraq being one of the most important centers in the thalassemia belt,[17] which has led many researchers to investigate the circumstances of gene flow. Thalassemia is the most common monogenic disease in the world and affects multiple ethnic and geographic groups with different prevalence in different locations.[18] The frequency of the -thalassemia gene is high and varies widely between different locations.[7] Throughout history, many populations have intermingled in Iraq. Invasion and migration processes play a key role in genetic mixing.[19] Other studies have shown that Iraqi populations are diverse.[20] Due to demographic heterogeneity, association studies between Turkey, Iran and Arab countries east of the Mediterranean have revealed a high number of thalassemia mutations.[21,22] The geographic and historical background of each site is reflected in the mutation distribution. Because of the high degree of consanguinity, especially in marriages between first cousins, there are a significant number of homozygotes who require monthly blood transfusions and chelation therapy. The high degree of endogamy in Arab communities is due to centuries-old sociocultural and religious traditions. However, no single mutation is restricted to Arabs, although different mutations exist in different Arab countries. Moreover, until recently, migration between Arab countries was widespread.[12]

Acknowledgements

To my wife Lamis Sahib, I would like to express my great appreciation and thanks to College of Medicine / University of Baghdad, Great thanks and gratitude should deeply be sent to members of Hematology unit in Thalassemia Centers in Wasit Government.

References

1. Abuzenadah AM, Hussein IM, Damanhouri GA, et al. Molecular basis of β-thalassemia in the western province of Saudi Arabia: identification of rare β-thalassemia mutations. Hemoglobin. 2011; 35(4): 346-57. doi: https://doi.org/10.3109/03630269.2011.588508.
2. Needs T, Gonzalez-Mosquera LF, Lynch DT. Beta Thalassemia. Treasure Island (FL): StatPearls Publishing; 2023. Available from: https://www.ncbi.nlm.nih.gov/books/NBK531481.
3. Al-Badran AI, Hassan MK, Washil AF. β-Thalassemia mutations among thalassemia major patients in Basrah Province-Iraq. Int J Curr Microbiol App Sci. 2016; 5: 448-57. doi: http://doi.org/10.20546/ijcmas.2016.505.047.
4. Weatherall DJ, Clegg JB. The Thalassaemia Syndromes. 4th ed. Oxford, U.K: Blackwell Science; 2001. doi: https://doi.org/10.1002/9780470696705.
5. Eissa AA, Kashmoola MA, Atroshi SD, Al-Allawi NA. Molecular Characterization of β-Thalassemia in Nineveh Province Illustrates the Relative Heterogeneity of Mutation Distributions in Northern Iraq. Indian J Hematol Blood Transfus. 2015; 31(2): 213-7. doi: https://doi.org/10.1007/s12288-014-0369-1.
6. Jalil T, Yousafzai YM, Rashid I, Ahmed S, Ali A, Fatima S, Ahmed J. Mutational Analysis Of Beta Thalassaemia By Multiplex Arms-Pcr In Khyber Pakhtunkhwa, Pakistan. J Ayub Med Coll Abbottabad. 2019; 31(1): 98-103. Available from: https://jamc.ayubmed.edu.pk/jamc/index.php/jamc/article/view/5403.
7. Akhavan-Niaki H, Derakhshandeh-Peykar P, Banihashemi A, et al. A comprehensive molecular characterization of beta thalassemia in a highly heterogeneous population. Blood Cells Mol Dis. 2011; 47(1): 29-32. doi: https://doi.org/10.1016/j.bcmd.2011.03.005.
8. da Guarda CC, Yahouédéhou S, Santiago RP, et al. Sickle cell disease: A distinction of two most frequent genotypes (HbSS and HbSC). PLoS One. 2020; 15(1): e0228399. doi: https://doi.org/10.1371/journal.pone.0228399.
9. Al-Allawi N, Al Allawi S, Jalal SD. Genetic epidemiology of hemoglobinopathies among Iraqi Kurds. J Community Genet. 2021; 12(1): 5-14. doi: https://doi.org/10.1007/s12687-020-00495-z.
10. Jomoui W, Panyasai S, Sripornsawan P, Tepakhan W. Revisiting and updating molecular epidemiology of α-thalassemia mutations in Thailand using MLPA and new multiplex gap-PCR for nine α-thalassemia deletion. Sci Rep. 2023; 13(1): 9850. doi: https://doi.org/10.1038/s41598-023-36840-8.
11. Hamid M, Dawoody Nejad L, Shariati G, Galehdari H, Saberi A, Mohammadi-Anaei M. The First Report of a 290-bp Deletion in β-Globin Gene in the South of Iran. Iran Biomed J. 2017; 21(2): 126-8. doi: https://doi.org/10.18869/acadpub.ibj.21.2.126.
12. Zahed L. The Spectrum of beta-Thalassemia Mutations in the Arab Populations. J Biomed Biotechnol. 2001; 1(3): 129-32. doi: https://doi.org/10.1155/s1110724301000298.
13. Rezaee AR, Banoei MM, Khalili E, Houshmand M. Beta-Thalassemia in Iran: new insight into the role of genetic admixture and migration. Scientific World Journal. 2012; 2012: 635183. doi: https://doi.org/10.1100/2012/635183.

14. Jiffri EH, Bogari N, Zidan KH, Teama S, Elhawary NA. Molecular updating of β-thalassemia mutations in the Upper Egyptian population. Hemoglobin. 2010; 34(6): 538-47. doi: https://doi.org/10.3109/03630269.2010.526440.
15. Al-Allawi NA, Al-Mousawi BM, Badi AI, Jalal SD. The spectrum of β-thalassemia mutations in Baghdad, Central Iraq. Hemoglobin. 2013; 37(5): 444-53. doi: https://doi.org/10.3109/03630269.2013.810641.
16. Moassas F, Nweder MS, Murad H. Hb Knossos (HBB: c.82G>T), β-globin CD 5 (-CT) (HBB: c.17_18delCT) and δ-globin CD 59 (-a) (HBD: c.179delA) mutations in a Syrian patient with β-thalassemia intermedia. BMC Pediatr. 2019; 19(1): 61. doi: https://doi.org/10.1186/s12887-019-1435-5.
17. Hamamy HA, Al-Allawi NA. Epidemiological profile of common haemoglobinopathies in Arab countries. J Community Genet. 2013; 4(2): 147-67. doi: https://doi.org/10.1007/s12687-012-0127-8.
18. Yang Z, Cui Q, Zhou W, Qiu L, Han B. Comparison of gene mutation spectrum of thalassemia in different regions of China and Southeast Asia. Mol Genet Genomic Med. 2019; 7(11): e1018. doi: https://doi.org/10.1002/mgg3.1018.
19. Chapin J, Cohen AR, Neufeld EJ, et al. An update on the US adult thalassaemia population: a report from the CDC thalassaemia treatment centres. Br J Haematol. 2022; 196(2): 380-89. doi: https://doi.org/10.1111/bjh.17920.
20. Saud AM. Molecular and biochemical study on β-thalassemia patients in Iraq. Doctoral dissertation, University of Baghdad; 2012. Available from: https://www.researchgate.net/publication/346040951.
21. el-Hazmi MA, Warsy AS, al-Swailem AR. The frequency of 14 beta-thalassemia mutations in the Arab populations. Hemoglobin. 1995; 19(6): 353-60. doi: https://doi.org/10.3109/03630269509005827.
22. Aliyeva G, Asadov C, Mammadova T, Gafarova S, Guliyeva Y, Abdulalimov E. Molecular and geographical heterogeneity of hemoglobinopathy mutations in Azerbaijanian populations. Ann Hum Genet. 2020; 84(3): 249-58. doi: https://doi.org/10.1111/ahg.12367.