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Comprehensive Molecular Diagnosis of α- and β-Thalassemia with Immunological Assessment of HBV/HCV Co-Infections in Transfusion among Iraqi Patients

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Abstract

Background: The alpha globin (*HBA1F*) and the beta globin (*HBB*) are the causative genes for thalassemia. The hepatitis B (HBV) and hepatitis C (HCV) are two viral infections that inflame the liver. **Objective:** The current study was planned to detect the thalassemia patients in both molecular and immunological aspects. **Methodology:** This case-control study that included 24 β-thalassemia patients and 10 healthy controls (both male and female, aged 10-35 years) who were admitted to thalassemia center at Babylon Governorate from March to December 2025. β-thalassemia was diagnosed by hemoglobin electrophoresis, and genomic DNA of the HBB and HBA1F genes was analyzed by conventional PCR. HBV and HCV infections were identified immunologically, and in silico molecular docking was used to investigate the interactions between BCL11A and hemoglobin inducers hydroxyurea and resveratrol. **Results:** Conventional PCR was used to confirm the presence of HBB gene in 100 % of the β-thalassemic patients and the absence of HBA1F gene in α-thalassemic patients. Patients showed elevated HbA2 (2.7–6.6%) and HbF (12.1%) versus normal ranges (2–3% and 0.8–2%), with >90% identified as carriers. Eleven samples (40.0%) were positive for HCV infection by immunological assays. The binding affinities of molecular docking with hydroxyurea and resveratrol were found to be strong with BCL11A, indicating increased expression of HbF and decreased severity of the disease. **Conclusion:** β-thalassemia showed a high hemoglobin F (HbF) levels by HB electrophoresis test, and over most of the thalassemia patients contract the hepatitis C virus while receiving blood transfusions. [*Bangladesh Journal of Infectious Diseases, December 2025;12(2):234-242*]

Keywords: α-thalassemia; β-thalassemia; *HBA1F*; HBB; HB electrophoresis rapid test; Immunological test; molecular docking

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Introduction

Thalassemia is inherited blood disorders which characterized by dysfunctional hemoglobin or abnormalities of a type of hemoglobin. A person's parents can pass on inherited diseases, such as thalassemia. The word "thalassemia" comes from the Greek word "thalassa," which means "sea," because this genetic disorder was first discovered in the Mediterranean region. Alpha-thalassemia and beta-thalassemia are the main types of thalassemia. Many genes cause thalassemia, and they are inherited in an autosomal recessive manner, but the most important genes are the *HBB* gene which is located on chromosome No.11, and the *HBA-1F* gene which is located on chromosome No.16, which both are well-thought-out the imperative gene mutations for the diagnosis of beta and alpha thalassemia. The sternness of the diseases is either determined by the loss of four α-globin genes or by the loss of two beta-globin genes.

There are two types of hemoglobin (Hb) diseases: thalassemia, which is usually due to defects in globin synthesis and regulation, and hemoglobinopathy, which results from structural defects of Hb, which are acquired and inherited forms in autosomal recessive[1,2]. Thalassemia can be classified as α, β, δβ, γδβ, δ, or γ depending on the type of globin chain or chain where production defects occur, e.g., α-thalassemia and synthesis of α-globin and the β-globin chains in disruption of β-thalassemia, in any case[3,4]. Thalassemia is more common in other parts of the world, such as Southeast Asia, the Indian subcontinent, and Africa. Additionally, the prevalence of thalassemia has recently increased due to increased immigration to places such as Australia, New Zealand, Northern Europe, and North America. An estimated 70,000 babies are born with various forms of thalassemia each year[5,6]. An estimated 7% of the world's population is expected to carry the thalassemia trait[7]. These stances a great risk to confront the spread of these diseases in the world and Iraq, and it is necessary to warn and prevent the marriage of relatives carrying the genes or any two carriers of the gene that causes thalassemia. That is common in Iraq, with a prevalence of 35.7 per 100,000. According to 2015 data, it is the most prevalent kind of inherited anemia. The two most common forms of thalassemia are α-thalassemia and β-thalassemia. Due to the lack of awareness and lack of understanding about the risks of this disease that the next generation of thalassemia patients may be unprotected, and also because of the lack of diagnosis for each person before marriage, which leads to an increase this disease a higher than 10% each year. This is a huge number for increasing this disease, and people must be sensitized to prevent the spread of thalassemia, especially in Iraq, where marriage between relatives is a major cause of thalassemia patients, and also the lack of genetic diagnosis for those who are about to marry[8].

Hemoglobin is also a common Hb alternative found in the Southeast Asian population. It has a relationship with a β-thalassemia phenotype, by way of people with thalassemia in this terrain being commonly found to have Hb. Transfusion-dependent and non-transfusion-dependent thalassemia are two new terminologies that are being used increasingly frequently in clinical settings, and all basic classifications fall into these two types depending on whether frequent blood transfusions are required or not and may lead to consequences such as venous thrombosis, HIV infection, osteoporosis, hypersplenism, and the chronic hepatitis (causes by infection with viruses that cause hepatitis B and/or C), etc.[9,10]. This study aimed to detect the various variants linked to thalassemia patients in the Babylon province as follows to determine whether the cause of thalassemia is related to *HBB* and *HBA1F* genes through verifying their presence in thalassemia patients. To conducting immunological tests to detect the presence of viral hepatitis types (B) and (C) in thalassemia patients who depend on blood transfusions every period.

Methodology

Study Population: A case-control study was conducted on the patients' group (24 β-thalassemia patients) who had already been diagnosed with thalassemia by symptoms and blood tests were randomly chosen at the Hematology Center, Department of Thalassemia, Babylon Hospital, for the period from March to December 2025 and their ages ranged from 10 to 35 years. During their routine blood test visits and, if required, prior to transfusion, they were asked to complete a specific questionnaire that the researchers requested and ten (10) healthy male and female individuals from Babylon's population who did not have hemoglobin-uropathy were randomized to be in a control group; their ages ranged from 10 to 35 years.

Blood Sampling: Five milliliters of blood were drawn by venous puncture from both patient's and the control group's, of which (Three milliliters) were drawn into EDTA tubes for DNA extraction, molecular assays, and immune-hepatitis diagnostic assays and (two milliliters) of blood were drawn into gel tubes. The samples were transferred using a cooling container within two to twenty-four hours, and kept in a refrigerator at 4ºC.

Laboratory Thalassemia Diagnosis: In thalassemia patients, the signs and symptoms appear as a result of both the disease itself and its treatment. Severe anemia, extramedullary hematopoiesis, and hemolytic appearances are related to the disease itself, while iron overload and oxidative stress manifestations occur as a result of frequent blood transfusion[11]. People with thalassemia have fewer healthy red blood cells, and have less hemoglobin than normal; those with α-thalassemia or β-thalassemia trait may have smaller-than-normal red blood cells, and a history of family and ethnicity may provide useful information in approaching the laboratory diagnosis of thalassemia[12]. The red cell morphology and their indices were used as hematological parameters for the laboratory identification of thalassemia patients, and the following tests were also used for the same purpose: Blood group, genetic testing by using conventional PCR, electrophoresis for hemoglobin to diagnose beta-thalassemia, and immune tests to detect the infection with hepatitis B and C.

Blood Group Test: A blood sample was taken from the thalassemia patients, and a drop of blood was placed on a glass slide, and the antigens A, B, and D were added to each blood drop and left for some minutes until the blood clotted[13].

Genomic DNA Extraction and Purification: Genomic DNA was extracted from preserved blood in EDTA using the Promega purification kit[14]. DNA concentration and purity were determined using a Nanodrop spectrophotometer, where the concentration was 200–250 ng/µl[15].

DNA Loading and Agarose Gel Electrophoresis: Six µl of DNA were mixed with one µl of loading medium and were carefully added to individual wells. For genomic DNA, electrophoresis at 70 volts continued for about 1 h, the product was then visualized using a UV transilluminator device[16].

Primer Designing and Dilution: Based on the previous research, the specific pair primers were designed for each gene using the primer designing tool in NCBI as follows: *HBA1-F* gene the expected size is 150 bp,
[F 5′-CCCACTGACCCTCTTCTCTG-3′]
[R 5′-CGGTATTTGGAGGTCAGCAC-3′]
and *HBB* gene the expected size is 190 bp.
[F 5′-TATCATGCCTCTTTGCACCA-3′]
[R 5′-AATCCAGCCTTATCCCAACC-3′]

The primers were synthesized at Bioneer/Korea and were provided in a lyophilized form. The primers were re-dissolved with deionized water according to the instructions of the manufacturer company to reach the final concentration (10 picomoles/*µl* of suspension). The nuclease-free water was added to *HBA1-F* forward and reverse primers (401.5 and 275 *µl*) and *HBB* forward and reverse primers (367 and 326 *µl*), all respectively.

Conventional Polymerase Chain Reaction (PCR): A polymerase chain reaction is typically used to diagnose thalassemia. Contamination between samples can occur during genomic DNA synthesis, and contaminated samples should be thrown away [17]. The traditional PCR test sought to find the α-globin and β-globin gene variants responsible for thalassemia syndrome. Using a 1 kb DNA ladder with DNA samples, the amplified DNA fragment by PCR was identified in order to evaluate whether or not the gene was present. The components of the PCR were 12 µl of master mix, 1 µl of forward primer, 1 µl of reverse primer, 5 µl of target DNA, and 6 µl of nuclease-free water for a total volume of 25 µl.

Conventional PCR Condition: A standard PCR condition[17] was used to determine the presence of *HBB* and *HBA1-F* genes in thalassemia patients (Table 1).

PCR Product Analysis: In each well, the ten microliters of the PCR product from the master mix were carefully added. The samples were exposed to electric shock for 90 minutes at 70 volts. DNA samples were run on a DNA ladder of 1000 bp to estimate the fragment size of each gene. UV light at a wavelength of 350 nm was used to visualize the DNA strands by a UV transilluminator device[18].

Table 1: PCR Condition for *HBB* and *HBA1-F* Genes

Step	**Temperatures** (°C)		**Time (minutes)**		**No. of Cycles**	
Genes	***HBB***	***HBA1-F***	***HBB***	***HBA1-F***	***HBB***	***HBA1-F***
Initial denaturation	95	95	3	2	1	1
Denaturation	95	95	1	1	35	30
Annealing	56	65	1	1		
Extension	72	72	1:30	1:30		
Final extension	72	72	7	5	1	1

HB Electrophoresis in Thalassemia: The hemoglobin electrophoresis with β-thalassemia trait usually has reduced or absent HbA, elevated levels of HbA2, and increased HbF. The distinguishing finding in beta-thalassemia is hemoglobin electrophoresis with the finding of elevated HgbA2 and F[19]. Both will be increased in the β-thalassemia trait without iron deficiency and will be normal or decreased in α-thalassemia and isolated iron deficiency anemia. The HB electrophoresis procedure[20], involved collecting the samples (blood); the samples were centrifuged at 1200 g for 5 min, following which the electrophoresis tank was filled out with TEB buffer, then the soaked cellulose acetate was soaked in the buffer solution for 5 minutes, then the well plate was filled with 5 *μl* of each diluted sample. Finally, the samples were covered with a glass slide to prevent evaporation and start the test.

Immunological Test of Viral Hepatitis: Hepatitis C and B Virus were diagnosed by investigating the virus antigens in the blood samples of the patients. The IgG antibodies were carried out using a rapid test called STREP to detect both HCV and HBV in thalassemia patients. According to the STREP kit, 10 *μl* of patient and control serum were put in the test strep for 10 minutes to allow the reaction to occur between the antibody and the antigen. The result appears as follows (where C is the control, and when it appears in the test strep, that means it is active and not damaged, while T means the infected patient with either the hepatitis C or B virus[10,21].

Molecular Docking Methods: The molecular docking analysis was conducted using AutoDock Vina to study the interaction between the BCL11A protein and selected fetal hemoglobin inducers (hydroxyurea and resveratrol). The three dimensional structure of BCL11A (PDB ID: 4YYE) was retrieved from the Protein Data Bank (https://www.rcsb.org/). Ligand structures were obtained from the PubChem database and prepared for docking using PyRx software. The all calculations for docking were performed under default parameters to predict binding affinities and interaction residues.

Statistical Analysis: Statistical analysis was performed using descriptive statistics. Quantitative data were expressed as mean ± standard deviation (Mean ± SD), whereas categorical variables were presented as frequencies and percentages. The obtained data were organized and analyzed using SPSS version 23.

Ethical Consideration: The ethics and protocol review were accepted by the ethics committee of the Department of Applied Biotechnology, College of Biotechnology, Al-Qasim Green University/Babylon/Iraq to perform this study (Number: 118. 210). Ethical clearance for sample collection was obtained from the ethics committee at the ministry of health/Iraq, and the individuals donating material gave their verbal and written consent.

Results

Distribution of samples by the age: The study was conducted from April to October 2025, and which include 24 female and male patients were randomly assigned in this study from Thalassemia Center/Babylon Hospitals (Table 2). The results showed that all patients in both genders and at all ages are affected by beta thalassemia; the age distribution of the thalassemia patients reflects the heterogeneity of β-thalassemia within different ages, which may indicate that β-thalassemia can occur in all age groups and in both gender types.

Table 2: Distribution of the Samples According to the Age Categories

Age Group	Patients	Control
10 to 20 years	3(12.3%)	3(30.0%)
21 to 30 years	14(58.4%)	6(60.0%)
30 to 35 years	7(29.3%)	1(10.0%)
Total	**24(100.0%)**	**10(100.0%)**

Hematological test (ABO blood group): The outcomes revealed that the patient blood groups were (A: 6, B: 5, AB: 3, and O: 10), while the control blood groups were (A: 2, B: 3, AB: 1, and O: 4. The lowest blood type was AB, while the highest blood type was O. All ABO groups were affected with beta-thalassemia, but the most affected blood groups were A and O (Figure I).

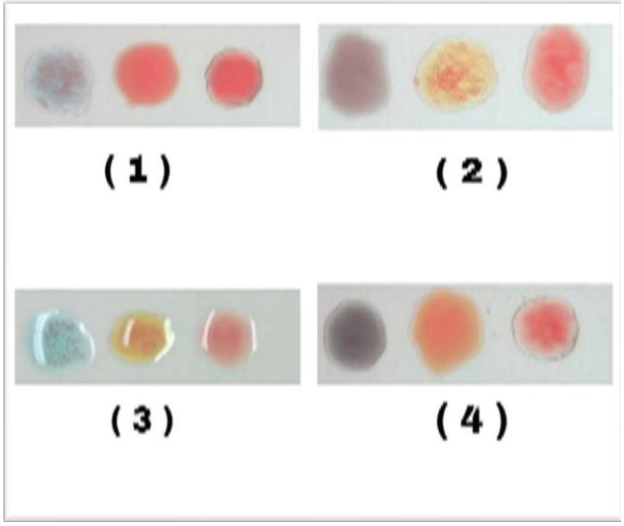

Figure I: ABO blood group. 1= (A Type), 2= (B Type), 3= (AB Type) and 4= (O Type)

Genomic DNA Extraction and Identification of β-Thalassemia: The kit from Promega purification was used to extract the genomic DNA from blood samples. A NanoDrop spectrophotometer at 260/280 nm was used to quantify the DNA (concentration and purity), which ranged from 58 to 142 ng/µl. The DNA product was verified by the horizontal gel electrophoresis in 1.0% agarose gel for 30 minutes at 75 volts; the product was then visualized by UV light, where the DNA appeared as compact bands (Figure II).

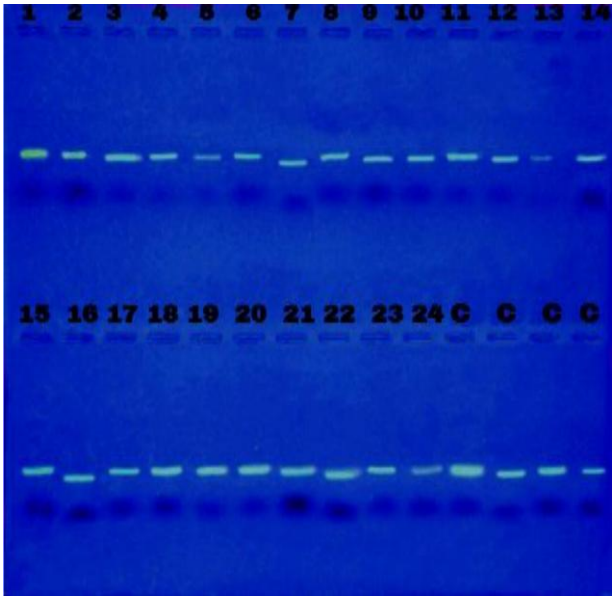

Figure II: Gel electrophoresis for the DNA of blood samples. Lines 1 to 24 represent the genomic DNA of patients, while the C lines represent the genomic DNA of the control samples

PCR Screening of HBB and HBA1-F genes in thalassemia patients: The outcomes showed that 100% of thalassemia sufferers had the HBB gene, which causes beta thalassemia. The expected product size of the HBB gene is 190 bp. Figure IIIA shows the result of the gel electrophoresis in 2% agarose gel at 80 volts for 60 minutes. The expected size of the HBA1-F gene is 150 bp. The samples were run using gel electrophoresis using 2% agarose gel at 80 volts for 1 hour, then a photo image was taken using the UV transilluminator at a wavelength of 254 nm. The findings did not show any band in all samples, meaning that all alpha thalassemia patients were not infected by the HBA1-F gene (Figure IIIB).

Electrophoresis of Hemoglobin in Patients with Thalassemia: The hemoglobin electrophoresis outcomes showed an increase in HbA2 and HbF in thalassemia patients compared with the control group. The HbA2 reached 6.6% (the normal percentage ranges from 2 to 3%), and the percentage of HbF reached 12.1% (the normal percentage is from 0.8 to 2%) (Table 3).

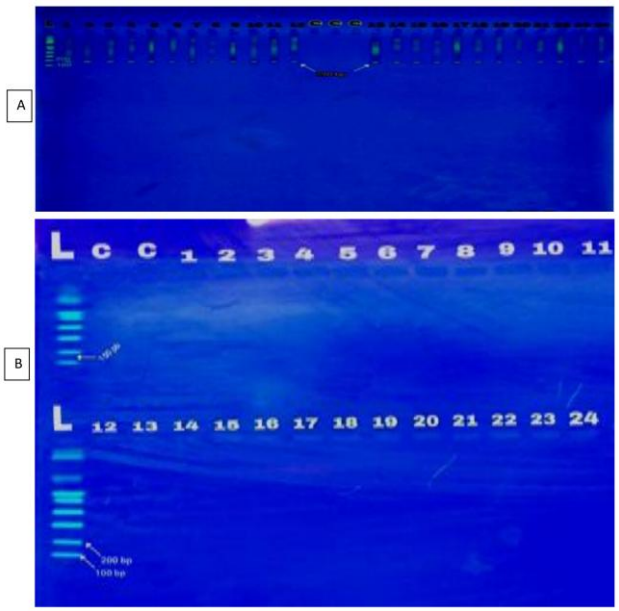

Figure III: Gel electrophoresis images. A. HBB gene. B. HBA1-F gene. Line L (1kb DNA ladder); lines 1 to 24 represent the HBB and HBA1-F genes in thalassemia patients; while C Lines Represent the Controls

Table 3: HB Electrophoresis of β-Thalassemia Patients (Percent)

HB Electrophoresis	Patient	Control
Hemoglobin A (HbA)	91.8±1.9	97.2±1.2
Hemoglobin A2 (HbA2)	4.8±1.2	1.5±0.3
Hemoglobin F (HbF)	4.1±1.5	0.5±0.2

Rapid Immuno-tests for HCV and HBV Diagnosis: The results of immunological tests showed that 11 (40%) of thalassemia patients (7 males and 4 females) out of 24 patients were infected with the hepatitis C virus sample, and no patient is infected with type B (Figure IV).

Molecular Docking Results: The docking results showed that all the tested compounds exhibited strong affinity toward the active site of BCL11A. Hydroxyurea displayed the highest binding affinity (–7.4 kcal/mol), followed by resveratrol (–6.8 kcal/mol) and curcumin (–6.3 kcal/mol). Hydrogen bonding and hydrophobic interactions were observed with key amino acid residues such as Arg290, Gln304, and Lys307. These findings suggest that hydroxyurea and resveratrol could inhibit BCL11A function and promote γ-globin gene activation, increasing fetal hemoglobin (HbF) production in β-thalassemia patients.

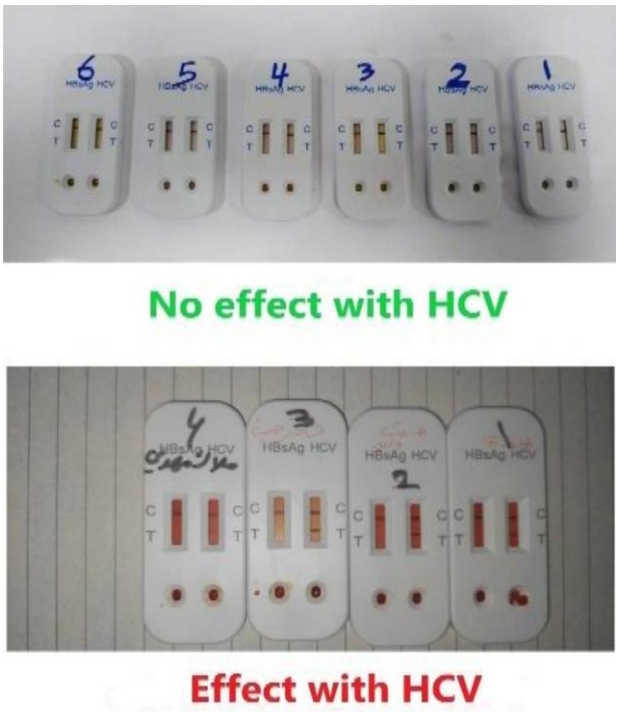

Figure IV: Immunological rapid tests (HCV, HBV). The C Line Stands for Control, while the T line stands for Patients Infected with HCV or HBV

The docking results showed that all the investigated drugs demonstrated significant affinity toward the active BCL11A's location. The greatest binding affinity was shown by hydroxyurea (–7.4 kcal/mol), followed by Curcumin (-6.3 kcal/mol) and resveratrol (-6.8 kcal/mol). Hydrogen bonding and hydrophobic interactions were detected with important amino acid residues such as Arg290, Gln304, and Lys307. These Research indicates that resveratrol and hydroxyurea may suppress BCL11A activity and increase γ- globin gene activation, boosting fetal hemoglobin (HbF) synthesis in β-thalassemia patients.

Alpha-thalassemia and beta-thalassemia are usually inherited in an autosomal recessive manner. This means that thalassemia does not affect sex hormones, but affects both men and women equally22. Minor beta-thalassemia is often detected incidentally during a routine complete blood count; mild anemia may be a symptom in patients without remarkable physical examinations. Beta thalassemia major (TM) is clinically diagnosed in

children under the age of two who have small cell anemia, moderate jaundice, and hepatic splenomegaly. Thalassemia intermedia presents with similar but milder clinical signs and manifests later in life. Occasionally carriers develop mild anemia, but most are asymptomatic[4,23].

Accordingly, there is no association between blood groups and phenotypes; this may be due to gene polymorphism of the ABO system since it is located on chromosome No.9, whereas the β-thalassemia gene is located on chromosome No.11[24,25]. The β globin gene can be downregulated due to a wide range of molecular lesions (type of damage in DNA structure), from point mutations to tiny deletions that just affect HBB to large deletions that affect the whole β globin cluster. While deletions are the primary cause of α-thalassemia, the majority of mutations producing β-thalassemia are non-delusional[26,27].

The degree of hepatic iron overload and the presence of chronic hepatitis C virus (HCV) infection are associated with the onset and severity of liver fibrosis, according to a multicenter study highly associated with Hepatitis is the main risk factor for liver fibrosis in transfusion-dependent thalassemia with a C-virus infection. Excess liver iron is now clearly recognized as a cofactor for the expansion of advanced fibrosis in patients with HCV infection. Even if it has clinical relevance, thalassemia-associated liver damage has been insufficiently characterized[28,29]. Most HCV-RNA-negative subjects with low iron burden did not develop hepatic fibrosis, but advanced fibrosis was common in HCV-RNA-positive patients with genotype 1 or 4 and iron overload of the appropriate chelation therapy. Usually in patients with chronic hepatitis C (CHC) thalassemia severe fibrosis growth capacity fatigue liver cirrhosis with HCV infection[18].

For the HB electrophoresis for thalassemia patients, this is because more than 90% of patients with beta-thalassemia were discovered to be carriers of the disease in the beta-thalassemia gene in addition to having high levels of both HbA2 and HbF4,13. Thalassemia major (TM) was diagnosed by hemoglobin electrophoresis, which often detects a raised HbF and HbA2[30]. In adults, these represent the typical fraction of hemoglobin molecules: 2% to 3% (0.02 to 0.03) for HbA, 0.8% to 2% (0.008 to 0.02) for HbF, and 95% to 98% (0.95 to 0.98) for HbA2. However, the typical HbF molecular levels in neonates and children are 50% to 80% (0.5 to 0.8) for neonates, 8% for 6-month-olds, and 1% to 2% for over 6 months. Hemoglobin levels that are too high or too low may mean they are sick with β-thalassemia.

Additionally, the interaction between the BCL11A repressor protein and recognized fetal hemoglobin inducers was investigated using in silico molecular docking research. The findings showed that hydroxyurea, resveratrol, and curcumin had substantial binding affinities to the active sites of BCL11A, indicating their possible involvement in preventing its suppressive effect on the expression of the γ-globin gene. By increasing HbF production, this inhibition may lessen the severity of β-thalassemia symptoms. Similar computational investigations have demonstrated that targeting BCL11A and similar transcriptional regulators may constitute an effective molecular strategy for reactivating fetal hemoglobin synthesis in β-thalassemia patients[8,12, 20,25].

Conclusion

The findings demonstrated that all thalassemia patients have beta-thalassemia due to the presence of the *HBB* gene. Alpha thalassemia patients, on the other hand, lacked the *HBA1F* gene. *HbA2* levels were greater in thalassemia patients than in the normal range. Similarly, the range of HbF were high in patients compared with control. Patients having high levels of HbA2 and HbF were identified with beta-thalassemia, and majority were a proved to be carriers of this condition. The immunological test findings of HBV and HCV in thalassemia patients conducted during blood transfusion revealed that a considerable proportion of patients were infected with the HCV virus.

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Conflict of Interest
The authors would like to state that there is no conflict of interest in this work.

Financial Disclosure
External funding was not obtained for this research.

Authors' contributions
Htma designed the study, wrote the manuscript, immunological

experiments, and analyzed the data; NFNA performed the molecular identification, wrote the molecular part, data analysis; GQH wrote and revised the manuscript with input from all authors, re-designing all tables and figures, and the corresponding author; MHA designed the study, wrote the immune part, performed the immunological experiments.

Data Availability
Any inquiries regarding supporting data availability of this study should be directed to the corresponding author and are available from the corresponding author on reasonable request.

Ethics Approval and Consent to Participate
The Institutional Review Board granted the study ethical approval. Since this was a prospective study, every study participant provided formal informed consent. Each method followed the appropriate rules and regulations.

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References

1. Lee YK, Kim HJ, Lee K, Park SH, Song SH, Seong MW, et al. Recent progress in laboratory diagnosis of thalassemia and hemoglobinopathy: a study by the Korean Red Blood Cell Disorder Working Party of the Korean Society of Hematology. Blood Res. 2019;54(1):17-22.
2. Idris ZS, Fatima SA, Hauwa SU, Aliyu DA, Bashiru I, Kastayal BS, et al. Thalassemia: pathophysiology, diagnosis, and advances in treatment. Thalass Rep. 2024;14(4):81-102.
3. Munkongdee T, Chen P, Winichagoon P, Fucharoen S, Paiboonsukwong P. Update in laboratory diagnosis of thalassemia. Front Mol Biosci. 2020;7:74.
4. Fasola FA, Babalola OA, Brown BJ, Odetunde A, Falusi AG, Olopade O. The effect of alpha thalassemia, HbF, and HbC on hematological parameters of sickle cell disease patients in Ibadan, Nigeria. Mediterr J Hematol Infect Dis. 2022;14(1):e2022001.
5. Motta I, Bou-Fakhredin R, Taher AT, Cappellini MD. Beta thalassemia: new therapeutic options beyond transfusion and iron chelation. Drugs. 2020;80(11):1053-1063
6. Dordevic A, Mrakovcic-Sutic I, Pavlovic S, Ugrin M, Roganovic J. Beta thalassemia syndromes: new insights. World J Clin Cases. 2025;13(10):100223.
7. Musharraf SG, Iqbal A, Ansari SH, Parveen S, Khan IA, Siddiqui AJ. β-thalassemia patients revealed a significant change of untargeted metabolites in comparison to healthy individuals. Sci Rep. 2017;7(1):42249.
8. Gong Y, Zhang X, Zhang Q, Zhang Y, Ye Y, Yu W, et al. A natural DNMT1 mutation elevates the fetal hemoglobin level via epigenetic derepression of the γ-globin gene in β-thalassemia. Blood. 2021;137(12):1652-1657.
9. Ahmadpanah M, Asadi Y, Haghighi M, Ghasemibasir H, Khanlarzadeh E, Brand S. In patients with minor beta-thalassemia, cognitive performance is related to length of education, but not to minor beta-thalassemia or hemoglobin levels. Iran J Psychiatry. 2019;14(1):47.
10. Ibrahim HF, Mahdi AS, Al Fatlawi SNM, Harrewee AO, Hameed RM. An immunological and molecular study to investigate the genes (β-globin and HBA1F) in patients with thalassemia in Najaf Governorate. J Prev Diagn Treat Strateg Med. 2024;3(2):85-98.
11. Fibach E, Dana M. Oxidative stress in β-thalassemia. Mol Diagn Ther. 2019;23(2):245-261.
12. Karponi G, Zogas N. Gene therapy for beta-thalassemia: updated perspectives. Appl Clin Genet. 2019;12:167-180.
13. Ali S, Mumtaz S, Shakir HA, Khan M, Tahir HM, Mumtaz S, et al. Current status of beta-thalassemia and its treatment strategies. Mol Genet Genomic Med. 2021;9(12):e1788.
14. Al-Mousawi HTM, Al-Hajjar QN. Immunological and molecular diagnosis of cytomegalovirus infection between aborted and pregnant women in Babylon city. Baghdad Sci J. 2021;18(2 Suppl):1086-1094.
15. Bunu SJ, Otele D, Alade T, Dodoru TRT. Determination of serum DNA purity among patients undergoing antiretroviral therapy using NanoDrop-1000 spectrophotometer and polymerase chain reaction. Biomed Biotechnol Res J. 2020;4(3):214-219ذ
16. Sadeghi MV, Mirghorbani M, Akbari R. β-thalassemia minor and renal tubular dysfunction: is there any association? BMC Nephrol. 2021;22(1):1-7.
17. Ryu J, Koh H, Lee HS, Lee H, Min J, Iwatsubo T, et al. Targeting microRNA-485-3p blocks Alzheimer's disease progression. Alzheimers Dement. 2021;17(Suppl 9):e057630.
18. McPherson RA, Pincus MR, editors. Henry's clinical diagnosis and management by laboratory methods. 24th ed. Philadelphia: Elsevier; 2021.
19. Zaghloul A, Al-Bukhari T, Bajuaifer N, Shalaby M, Al-Pakistani H, Halawani SH, et al. Introduction of new formulas and evaluation of the previous red blood cell indices and formulas in the differentiation between beta thalassemia trait and iron deficiency anemia in the Makkah region. Hematology. 2016;21(6):351-358.
20. Al-Allawi NA, Puehringer H, Raheem RA, Oberkanins C. Genetic modifiers in β-thalassemia intermedia: a study on 102

Iraqi Arab patients. Genet Test Mol Biomarkers. 2015;19(5):242-247.
21. Alsahib SA, Alhussein AA, Jameel YM. Estimation of CD8 parameter level in thalassemia patients. J Technique. 2022;4(Special Issue):110-115.
22. Yang CHT, Yoo ER, Ahmed A. The role of direct-acting antivirals in the treatment of children with chronic hepatitis C. J Clin Transl Hepatol. 2017;5(1):59.
23. Al-Akhras A, Badr M, El-Safy U, Kohne E, Hassan T, Abdelrahman H, et al. Impact of genotype on endocrinal complications in β-thalassemia patients. Biomed Rep. 2016;4(6):728-736.
24. Wahed A, Quesada A, Dasgupta A. Hematology and coagulation. 2nd ed. Elsevier; 2019.
25. Rashid NM, Sultan AA. Assessing the impact of HBB gene polymorphisms on the risk of beta thalassemia in the population of Diyala Governorate, Iraq. Iraq Med J. 2024;8(2):35-39
26. Nandakumar SK, Ulirsch JC, Sankaran VG. Advances in understanding erythropoiesis: evolving perspectives. Br J Haematol. 2016;173(2):206-218.
27. Arwa HMA. Evaluation of some heart enzymes and iron levels in β-thalassemia patients in Thi-Qar City, Iraq. Baghdad Sci J. 2024;21(6):2007-2016.
28. Rösler R, Abbas MD, Papp T, Marschang RE. Detection of antibodies against paramyxoviruses in tortoises. J Zoo Wildl Med. 2013;44(2):333-339.
29. Sleiman J, Tarhini A, Bou-Fakhredin R, Saliba AN, Cappellini MD, Taher AT. Non-transfusion-dependent thalassemia: an update on complications and management. Int J Mol Sci. 2018;19(1):182
30. Gupta A. Thalassemia major. In: Decision making through problem based learning in hematology: a step-by-step approach in patients with anemia. Singapore: Springer Nature; 2024. p.35-51