Review Article

Molecular Assessment of Minimal Residual Disease in Iranian Pediatric Acute Lymphoblastic Leukemia Patients

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Abstract

Acute lymphoblastic leukemia (ALL) is the most common hematologic malignancy in children. TAL1 dysregulation drives 40–60% of T-ALL cases, but its utility as a minimal residual disease (MRD) biomarker remains controversial. We evaluated TAL1 expression in pediatric T-ALL patients. This case-control study analyzed TAL1 expression via RT-PCR in 50 T-ALL patients (new diagnosis, post-induction, relapse, aged 0–14 years) and 30 controls. SIL-TAL1 fusion status was assessed by using real-time polymerase chain reaction (RT-PCR). The study also assessed correlations between TAL1 expression and the patients' demographic and laboratory characteristics. GAPDH served as the internal control gene. TAL1 was overexpressed in new diagnoses (5.2-fold vs. controls; $p<0.001$) and relapse (3.1-fold vs. post-induction; $p<0.005$). SIL-TAL1+ patients had higher relapse rates (HR=2.3; 95% CI:1.1–4.8). TAL1 overexpression correlates with disease activity in T-ALL, but SIL-TAL1 fusion may better predict relapse. Larger validation studies are needed. Or (These findings suggest that TAL1 overexpression is associated with disease burden and relapse, supporting its potential as a genetic MRD biomarker in Iranian pediatric ALL patients).

Keywords: Pediatric T-ALL, Minimal residual disease, RT-PCR, TAL1, SIL-TAL1 fusion, Relapse

Introduction

Acute lymphoblastic leukemia (ALL) is the most common pediatric cancer, accounting for approximately 30% of all childhood malignancies. It arises from the clonal expansion of B- or T-lymphoblasts in the bone marrow (BM). Despite significant advancements in treatment, children categorized as high-risk continue to face poor outcomes. High-risk subgroups include molecular subtypes such as BCR-ABL fusion, cytogenetic abnormalities like hypodiploidy, and immunophenotypic subtypes such as T-cell ALL or B-cell ALL with BCR-ABL fusion [1, 2]. Clinical trials have achieved remarkable progress, with 5-year event-free survival (EFS) rates exceeding 85% and overall survival (OS) rates surpassing 90% for B-cell ALL. However, outcomes for T-cell ALL remain consistently lower by 5–10% across most studies [1, 2].

Several risk factors influence treatment outcomes, including white blood cell count, patient age, cytogenetic findings, immune phenotype, and early response to corticosteroids. Tailoring treatment protocols based on these factors can reduce drug toxicity in low-risk patients while improving outcomes for high-risk cases. Molecular investigations into disease progression have unveiled novel therapeutic strategies, advancing diagnosis and treatment approaches. Key genes involved in epigenetics, immune system regulation, transcription factors, tyrosine kinase signaling, and apoptosis pathways play critical roles in leukemia pathogenesis [3]. According to GLOBOCAN statistics from 2022, ALL exhibits a high survival rate in children and is more prevalent among boys aged 0–14 years [4]. While approximately 90% of patients achieve complete remission, a significant proportion fail to respond to treatment, leading to relapse or mortality in some cases [5].

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Despite these advances, relapse remains the most significant cause of treatment failure in pediatric ALL, resulting in poor prognosis and contributing to ALL being one of the leading causes of cancer-related deaths in children [8]. Relapse risk stratification at diagnosis classifies low- and intermediate-risk patients with a 5-year EFS rate of approximately 90%. In contrast, high-risk patients achieve an 80% survival rate with intensified therapy [9]. Minimal residual disease (MRD) assessment has emerged as a critical prognostic factor, enabling the prediction of relapse and guiding treatment decisions. MRD serves as a sensitive diagnostic tool, detecting one leukemic blast cell in a background of one million cells [8]. Flow cytometry and reverse transcriptase polymerase chain reaction (RT-PCR) are widely used techniques for MRD monitoring, with RT-PCR regarded as the gold standard in clinical laboratories [10, 11].

The biology of relapsed ALL is characterized by considerable genetic heterogeneity at both chromosomal and single-gene levels. Driver mutations, chromosomal aberrations, and deregulated signaling interactions between leukemia cells and the immune microenvironment contribute to disease progression, particularly in T-cell ALL (T-ALL). Recent insights into T-ALL biology using modern genomic techniques have identified recurrent lesions that cluster into targetable pathways, including Notch, Jak/Stat, PI3K/Akt/mTOR, and MAPK. With contemporary chemotherapy, outcomes for de novo T-ALL have improved significantly, approaching those observed in B-ALL, with approximately 85% 5-year EFS. However, salvage rates remain poor, with less than 25% EFS and OS rates for relapsed disease [13].

Despite these strides, challenges persist. Targeted therapies and immunotherapies have shown limited success in T-ALL, unlike in Philadelphia chromosome-positive B-ALL, where tyrosine kinase inhibitors (TKIs) such as imatinib and dasatinib have markedly improved survival [13]. The lack of effective targeted treatments for T-ALL underscores the need for further research into its molecular mechanisms. Among the genes implicated in ALL biogenesis, TAL1 stands out due to its critical role in hematopoietic development and leukemogenesis. TAL1 is overexpressed in 60% of pediatric ALL cases, often due to chromosomal translocations or alterations in its regulatory regions, even in the absence of structural rearrangements [16, 17]. Its interaction with genes such as ABL1, STIL, and CDKN2A highlights its central role in leukemia progression [18].

To address these gaps, this study focuses on evaluating TAL1 expression profiles in Iranian pediatric ALL patients, a topic with limited prior research. We analyzed TAL1 expression at three critical time points—diagnosis, post-induction, and relapse—and compared these profiles to a control group of healthy individuals. Our findings align with recent studies from other geographic regions, suggesting that TAL1 expression may serve as a potential biomarker for predicting MRD levels and relapse risk. This research aims to enhance understanding of TAL1's role in ALL biogenesis and inform treatment protocol management for improved outcomes [19].

Materials and Methods

Peripheral blood (PB) specimens were collected from 50 pediatric patients with acute lymphoblastic leukemia (ALL) admitted to Mofid Children's Hospital Research Center and Children's Medical Center in Tehran, Iran. Written informed consent was obtained from all participants or their guardians prior to sample collection. The study was approved by the Ethics Committee of the College of Sciences at the University of Tehran (IR.UT.SCIENCE.REC.1401.001).

Newly diagnosed ALL patients received standard induction chemotherapy consisting of vincristine, glucocorticoids, and L-asparaginase, with or without daunorubicin. Follow-up sampling was conducted for all 50 patients during the post-induction phase after confirmation of complete remission through para-clinical evaluations **(Figure 3)**. Additionally, PB samples were collected from 20 patients who relapsed during the study period. As a control group, peripheral blood samples were also obtained from 30 healthy individuals.

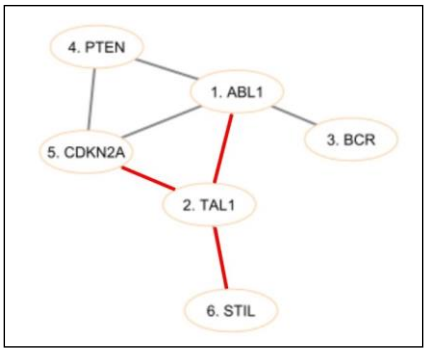

Figure 1.TAL1 role in Pediatric ALL Biogenesis (Cystoscope Analysis)

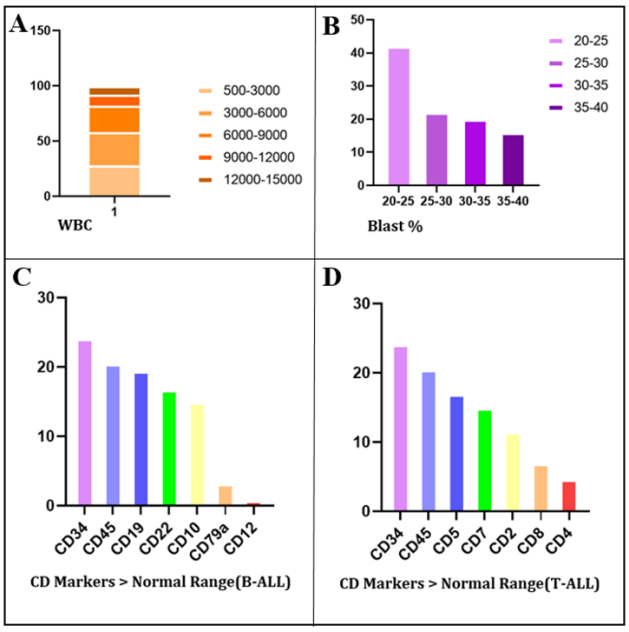

Figure 2. Paraclinical Tests Results (New Case Time Point) (A. WBC, B. Blast Percentage, C. B-ALL CD Markers, D. T-ALL CD Markers)

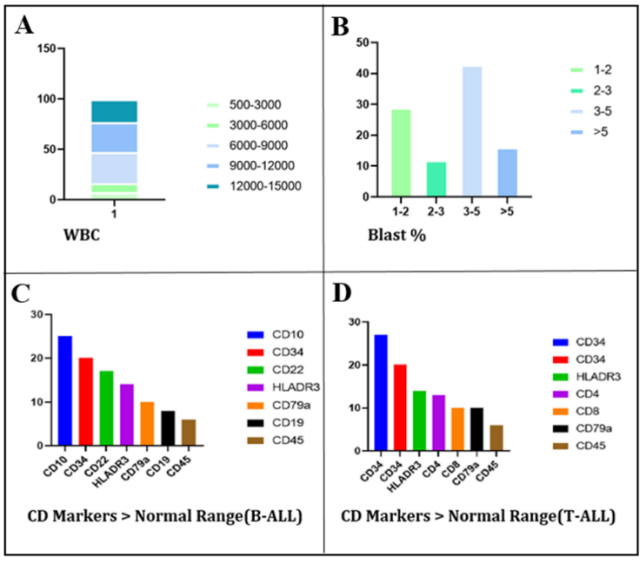

Figure 3. Paraclinical Tests Results (Post-Induction Time Point)

(**A**. WBC, **B.** Blast Percentage, **C.** B-ALL CD Markers, **D.** T-ALL CD Markers)

Specimen collection

Peripheral blood (PB) samples were collected from pediatric patients diagnosed with acute lymphoblastic leukemia (ALL). Diagnostic test results revealed the following patient characteristics: 38.9% of patients did not belong to any known molecular subgroup, while 24.1% were classified as B-cell ALL (B-ALL), 18.5% as Philadelphia chromosome-like ALL (Ph-like ALL), and 18.5% as T-cell ALL (T-ALL). Among the cohort, 28% of patients had a white blood cell (WBC) count below 3,000 K/µL, while 18% had a WBC count of 9,000 K/µL. Additionally, 42% of patients exhibited blast counts exceeding 20%, with 16% showing blast counts above 35%. Flow cytometry analysis of CD markers revealed notable deviations from normal expression levels. In 44.7% of cases, CD34 and CD45 were overexpressed. For B-ALL cases, CD19, CD22, and CD10 showed elevated expression in 41.3% of patients, while in T-ALL cases, CD5, CD7, CD2, and CD4 were overexpressed in 48.5% of patients **(Figure 2)**. Peripheral blood samples were collected in EDTA-containing tubes and stored at -20°C until further processing. After the collection period, samples were thawed and washed twice using phosphate-buffered saline (PBS, pH 7.4, 0.15 M, Gibco, UK). The resulting cell pellets were lysed directly using TriPure Isolation Reagent (Roche, Germany).

Cytogenetic analysis

Bone marrow aspirates from patients were directly cultured and harvested using standard cytogenetic methods. G-banding and fluorescence in situ hybridization (FISH), when necessary, were performed on each bone marrow sample to detect all cytogenetic abnormalities in accordance with an in-house validated protocol. Chromosomal aberrations were interpreted following the guidelines outlined in the International System for Human Cytogenetic Nomenclature (ISCN 2020) [20]. The identified chromosomal abnormalities are listed in **Table 2**.

Table 1. Genetic Alterations in Pediatric ALL

B-ALL Genetic Alterations	T-ALL Genetic Alterations	B and T ALL Genetic Alterations
ETV6–RUNX fusion	Transcription Factor Oncogenes	KMT2A Rearrangement
Aneuploidy	Signaling Pathway Notch Abnormally	BCR–ABL1*
BCR-ABL Fusion	Deregulation of Functional Pathways	Somatic Translocations(MLL)
TCF3–PBX1 and HLF fusion genes	PI3K/AKT/mTOR Signaling	KMT2A Rearrangements
Other	Other	-----

*BCR–ABL1 fusions are common in B-cell ALL, and in T-cell ALL, ABL1 can form fusions with BCR, NUP214, and EML1; 34.

Table 2. Cytogenetic Abnormalities Identified at Diagnosis and Post-Induction

#	Chromosomal Abnormality	Percentage(New Case)	Percentage(Post-Induction)
1	t (12;21)(p13;q22)/ETV6-RUNX1	13.3%	7
2	t (4;11) (q21; q23)/KMT2A-AFF1	10%	6
3	+Philadelphia Chromosome	8.3%	5
4	t (1;19) (q23; p13)/TCF3-PBX1	6.7%	3
5	t (9;22) (q34; q11)/BCR-ABL	6.7%	4.7
6	t (1;7) (p32;q34)/BTF3L-RAF	5.2%	1.3
7	del (11) (q23)/MLL	5.2%	2.3
8	t (7;10) (q34; q24)/TRB-HOX11	3.35%	2.7
9	t (5;14) (q35;q11)/RANBP17-TRD	3.35%	2.5
10	t (7;14) (p15; q32)/TRC-TCL1A	3.3%	1.4
11	t (8;14) (q24; q32)/MYC-IGH	1.6%	0.6
Total	Abnormal Karyotype	67%	36.5
#	Normal Karyotype	Percentage	
1	46, XX or 46, XY	33%	63.5

Total RNA extraction and cDNA synthesis

Total RNA was isolated from frozen peripheral blood (PB) samples using RNX-Plus Isolation Reagent (RNX-Plus/Sinaclon/Iran), following the manufacturer's protocol [21]. The quantity and quality of the RNA samples were

assessed by measuring absorbance at 260/280 nm wavelengths using a NanoDrop spectrophotometer. Subsequently, 1 µg of total RNA was reverse-transcribed into complementary DNA (cDNA) using the SMOBIO First Strand cDNA Synthesis Kit (SMBIO Technology, Belarus) [22]. The synthesized cDNA was stored at -20°C for further analysis.

RT-PCR Analysis

To assess the relative quantity of mRNA transcripts, real-time PCR (RT-PCR) was performed using a StepOne Plus Real-Time PCR System (Bio-Rad, USA) with the SYBR Green assay in duplicate. The cycling conditions included an initial denaturation step at 95°C for 10 minutes, followed by 40 cycles of 95°C for 15 seconds and 61°C (combined annealing/extension) for 1 minute. A melting curve analysis was subsequently performed to confirm primer specificity for each target gene. To determine the efficiency of the RT-PCR reactions, a standard curve was generated using a serial dilution (5-fold dilutions) of cDNA samples.

All reactions were carried out in a final volume of 20 µl, consisting of 10 µl qPCR Master Mix (Qiagen), 2 µl (200 ng/µl) of cDNA, 1 µl of each primer, and 6 µl of nuclease-free water (ddH_2O). The expression levels of TAL1 were normalized to GAPDH, a housekeeping gene recommended for such analyses by the American Society of Clinical Oncology (ASCO) [23]. Relative quantification was performed using the $2^{-\Delta\Delta Ct}$ method [24, 25]. Primers were designed using the publicly available Primer3 software [26, 27], and their details are provided in **Table 3**.

Table 3. The list of Primers (5' -> 3')

Primer Name	TAL1
Forward	ACAACCGAGTGAAGAGGAGACC: 22
Reverse	TCACATTCTGCTGCCGCCAT: 20
Primer Name	**GAPDH**
Forward	CGGATTTGGTCGTATTGGGC: 20
Reverse	TTCTCAGCCTTGACGGTGCCATG: 23

Statistical analysis

The Mann-Whitney U test was used to compare the expression levels of TAL1 and GAPDH between healthy individuals and pediatric patients with acute lymphoblastic leukemia (ALL). This non-parametric test was chosen due to the non-normal distribution of the data. The same test was also applied to compare expression levels at two critical time points: the beginning of treatment and post-induction. Additionally, the Mann-Whitney U test was used to compare the two ALL subgroups (B-cell ALL and T-cell ALL) and to evaluate expression levels in patients at relapse, specifically for PDGFRB and GAPDH. A P -value of less than 0.005 was considered statistically significant. All statistical analyses were performed using SPSS version 20 (SPSS, Chicago, IL, USA) and REST 2009 software.

Patients Follow-Up

Patient follow-up was conducted through direct communication with both patients and their parents. All participants were enrolled in the study after receiving genetic counseling and providing informed consent. To support the families, we offered financial and psychological assistance whenever possible. Additionally, we introduced the children to basic genetic concepts in simple, accessible language to help them understand the importance of the study. To make their experience more positive, we provided small gifts, such as handmade dolls, every Saturday, aiming to create a supportive and encouraging environment throughout their treatment journey. Following the induction phase, peripheral blood samples were collected again, and molecular tests were performed to assess minimal residual disease (MRD) at the genetic level.

Results and Discussion

I. Patient Characteristics at Diagnosis

a. Para-clinical Findings

Upon initial hematological screening and para-clinical testing of peripheral blood (PB) samples from the 50 pediatric ALL patients, leukemia was confirmed. The distribution of molecular and immunophenotypic subgroups within the cohort was as follows: 38.9% were not classified into any known molecular subgroup, 24.1% were B-cell ALL (B-ALL), 18.5% were Philadelphia chromosome-like ALL (Ph-like ALL), and 18.5% were T-cell ALL (T-ALL). At diagnosis, 28% of patients presented with a white blood cell (WBC) count below 3,000 K/µL, while 18% had a WBC count exceeding 9,000 K/µL. Blast counts were above 20% in 42% of patients, with 16% exhibiting counts greater than 35%. Flow cytometry analysis revealed overexpression of CD34 and CD45 in 44.7% of cases. In B-ALL patients, CD19, CD22, and CD10 showed elevated expression in 41.3% of cases, whereas T-ALL patients exhibited overexpression of CD5, CD7, CD2, and CD4 in 48.5% of cases **(Figure 2)**.

b. Cytogenetic Analysis

Cytogenetic analysis of bone marrow (BM) samples identified various chromosomal abnormalities **(Table 2)**. The most frequent rearrangement was t(12;21) (p13;q22), observed in 13.3% of patients, while t(8;14)(q24;q32) was the least frequent, occurring in 1.6% of cases. Notably, one patient with T-ALL/LBL was diagnosed with t(9;22);BCR-ABL1 (Ph+). Approximately 33% of patients presented with a normal karyotype. Based on cytogenetic findings, patients were categorized into B-cell and T-cell ALL subgroups. The average age of the cohort was 8 years, with a gender distribution of 40% female and 60% male. Statistical analysis revealed no significant correlation between gender and the expression levels of TAL1 or GAPDH at any of the studied time points (data not shown).

c. TAL1 and GAPDH Expression Analysis by RT-PCR

Quantitative assessment of TAL1 and GAPDH mRNA expression levels at diagnosis was performed using RT-PCR. The primers utilized for this analysis are detailed in **Table 4**. The relative expression levels of TAL1, normalized to the housekeeping gene GAPDH, are presented in **Figure 4**. Statistical analysis using the Mann-Whitney U test revealed a significantly higher expression of TAL1 in newly diagnosed ALL patients compared to the healthy control group ($P < 0.005$). Furthermore, when comparing the B-ALL and T-ALL subgroups at diagnosis, TAL1 expression levels were significantly elevated in the T-ALL subgroup compared to the B-ALL subgroup ($P < 0.005$) **(Figure 4)**.

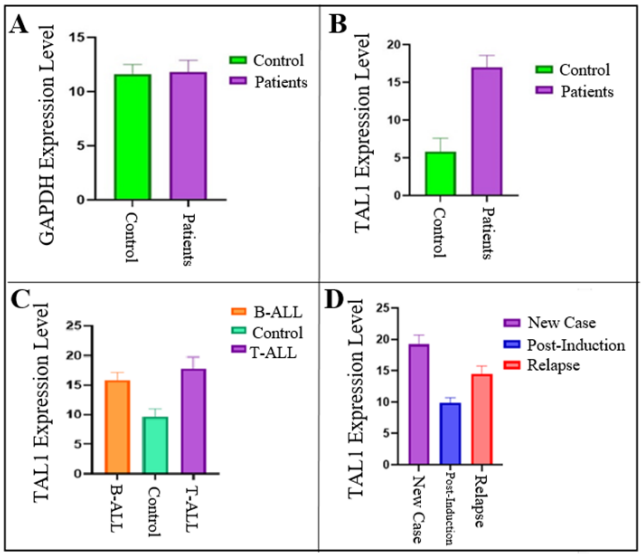

Figure 4. Expression level of TAL1 and GAPDH (Fold Change)
(**A**. Control and Patient. GAPDH, **B.** Control and Patient. TAL1, **C.** B and T-ALL. TAL1, **D.** Different Time Points. TAL1).

II. Patient Characteristics and TAL1 Expression Post-Induction

Following the induction phase of chemotherapy (28-33 days), para-clinical tests and cytogenetic analysis were repeated to confirm complete hematological remission **(Figure 3)**. Post-induction evaluation showed white blood cell (WBC) counts ranging from 6,000 to 15,000 K/µL, with blast counts reduced to 2-5%. Flow cytometry analysis indicated a normalization of CD34 and CD45 expression in both B-ALL and T-ALL patient subgroups **(Figure 3)**. Cytogenetic analysis at this time point revealed a persistence of t(12;21)(p13;q22) as the most frequent abnormality (7%), while t(8;14)(q24;q32) remained the least frequent (0.6%). The proportion of patients with a normal karyotype increased to 63.5% **(Table 2)**.

RT-PCR analysis of TAL1 expression in post-induction PB samples demonstrated a significant decrease in TAL1 expression levels compared to the diagnostic time point in the overall ALL cohort ($P < 0.005$) **(Figure 4)**. Notably, this reduction was more pronounced in the T-ALL subgroup. However, TAL1 expression in both B-ALL and T-ALL subgroups remained significantly higher than in the healthy control group ($P < 0.005$) **(Figure 4)**.

III. Minimal Residual Disease (MRD) Assessment and TAL1 Expression

To assess minimal residual disease (MRD) at the molecular level, TAL1 expression was evaluated again by RT-PCR in post-induction samples. Elevated TAL1 expression levels post-induction was associated with higher MRD burden. Specifically, patients with higher TAL1 expression post-induction were more likely to experience relapse during the follow-up period (this observation will be further quantified and statistically analyzed in the subsequent sections/discussion).

Comparative analysis of TAL1 expression between the diagnostic and post-induction time points revealed a significant decrease in both B-ALL and T-ALL subgroups following treatment. However, the T-ALL subgroup exhibited significantly higher TAL1 expression at diagnosis and, despite the reduction post-induction, maintained a higher level of expression compared to the B-ALL subgroup **(Figure 4)**.

IV. TAL1 Expression at Relapse

In the 20 patients who relapsed during the study period, TAL1 expression levels in PB samples collected at the time of relapse were significantly higher compared to the post-induction remission samples ($P < 0.005$) and, in many cases, approached or even exceeded the levels observed at initial diagnosis **(Figure 4)**. This increase in TAL1 expression at relapse was observed in both B-ALL and T-ALL patients, suggesting a potential role for TAL1 upregulation in disease recurrence.

Interpretation and Recommendations

The observed higher TAL1 expression in T-ALL patients at diagnosis, its subsequent decrease post-induction, and its resurgence at relapse suggest that TAL1 expression levels may serve as a valuable biomarker for disease monitoring and MRD assessment, particularly in the T-ALL subgroup. The persistent elevation of TAL1 expression post-induction compared to healthy controls indicates the potential for this gene to be a marker of residual disease. The significant increase in TAL1 expression at relapse further supports its involvement in disease progression and recurrence in both B-ALL and T-ALL. Therefore, we recommend the inclusion of TAL1 expression analysis in the routine monitoring of pediatric ALL patients, especially those with T-ALL, at diagnosis and during MRD assessment post-induction to potentially predict relapse risk and guide treatment strategies. Further investigation with a larger cohort and longer follow-up is warranted to validate these findings and explore the potential of TAL1 as a therapeutic target.

Leukemia, with an increasing prevalence in Iran over the last decade, represents a significant health concern [Reference

30]. Accurate diagnostic and prognostic methods are crucial for effective management of this disease, where epigenetic factors and immune responses are increasingly recognized for their role in the Iranian population. Population-based genetic profiling studies in Iran have identified potential prognostic biomarkers with diagnostic utility [Reference 30], highlighting the importance of understanding the specific molecular landscape of leukemia within this population.

Acute lymphoblastic leukemia (ALL), characterized by the impaired maturation and differentiation of blood cells in the bone marrow, is the most common childhood cancer. Advances in molecular biology have led to a refined classification of ALL based on genetic alterations, including tumor suppressor genes, oncogenes, and fusion genes resulting from chromosomal translocations, which hold significance for early detection.

Both environmental and genetic factors contribute to cancer development and progression. Lifestyle changes and geographical variations can influence epidemiology and the microbiome, potentially impacting cancer prevalence. Iran's diverse climate and ethnic groups present a unique opportunity to investigate the role of the microbiome in ALL biogenesis, and our study's collection of samples from various geographical regions aims to facilitate future research in this area.

The transformation of normal bone marrow cells into leukemic cells is often driven by mutations in oncogenes (promoting cell growth and survival) and tumor suppressor genes (regulating cell growth and division). Specific chromosomal rearrangements are common in leukemia and serve as important prognostic indicators. Cytogenetic analysis, including G-banding and FISH, remains a cornerstone of T-ALL diagnosis, identifying recurrent chromosomal abnormalities and guiding diagnostic frameworks [Reference 32, 33]. While certain translocations like t(12;21), t(4;11), t(1;19), and +Ph are well-established in ALL, t(9;22)(BCR::ABL1) is often a secondary abnormality in T-ALL, leading to ABL1 overexpression **(Table 2)**. Notably, a significant proportion (25-30%) of T-ALL cases lack established cytogenetic abnormalities [Reference 32, 33], underscoring the need for complementary molecular markers.

The economic burden of cancer management on healthcare systems, particularly for childhood ALL, necessitates cost-effective strategies. MRD evaluation, by informing treatment intensity and duration, has the potential to reduce the overall cost of care. Targeted therapies, directed at specific genetic alterations, also offer promise in reducing treatment costs by employing more selective and potentially less toxic agents. The prognostic significance of MRD assessment, using techniques like flow cytometry and RT-PCR, is well-established, with negative MRD correlating with favorable outcomes and complete remission [Reference 32]. The increasing sensitivity of MRD detection methods is paving the way for personalized medicine in ALL. Approximately 30% of ALL patients are at risk of relapse [Reference 34], making accurate MRD assessment crucial for identifying this high-risk group. While molecular subgroups are better defined in B-cell precursor ALL, ongoing research aims to refine risk stratification in T-lineage ALL based on transcriptomic, epigenetic, and mutational profiles [Reference 35]. European guidelines emphasize the importance of post-induction MRD evaluation for ALL patients, and new therapies like monoclonal antibodies and engineered T cells further underscore the need for robust MRD monitoring to guide personalized treatment strategies.

In this study, we focused on *TAL1* expression as a potential novel biomarker for MRD assessment in Iranian pediatric ALL patients using RT-PCR. *TAL1*, a transcription factor critical for hematopoiesis and implicated in leukemogenesis [Reference 37], has been shown to be overexpressed in a significant proportion (60%) of pediatric ALL cases at diagnosis [Reference 38]. Chromosomal rearrangements involving *TAL1*, such as its addition to *STIL*, are common in T-ALL and are associated with poor treatment outcomes [Reference 38]. Even in the absence of structural rearrangements, altered *TAL1* expression can occur due to various genetic and epigenetic mechanisms [Reference 38].

Our findings reveal a significantly higher expression of *TAL1* in newly diagnosed ALL patients compared to healthy controls, with even higher levels observed in the T-ALL subgroup **(Figure 4)**. Following induction chemotherapy, *TAL1* expression significantly decreased in both B-ALL and T-ALL patients, correlating with the achievement of hematological remission. Importantly, we observed that elevated *TAL1* expression post-induction was associated with a higher likelihood of relapse, suggesting its potential as a marker for residual disease. Furthermore, *TAL1* expression was markedly increased at the time of relapse in both B-ALL and T-ALL patients, indicating its potential involvement in disease recurrence.

These results align with recent international studies highlighting the role of *TAL1* as a biomarker in ALL [Reference 50]. The consistent pattern of *TAL1* expression across diagnosis, post-induction, and relapse suggests its utility in monitoring disease burden and predicting relapse risk, particularly in T-ALL. Given *TAL1*'s critical role in hematopoietic development and its frequent dysregulation in ALL, our findings support its potential as a valuable biomarker for MRD evaluation and a potential therapeutic target.

The genetic evaluation of MRD using RT-PCR and novel biomarkers like *TAL1* can contribute to more tailored treatment protocols and a more efficient allocation of healthcare resources in leukemia management. Our study underscores the importance of investigating population-specific biomarkers in ALL and recommends the integration of *TAL1* expression analysis into the routine monitoring of pediatric ALL patients in Iran, especially those with T-ALL, to potentially improve risk stratification and guide treatment decisions. Further research with larger cohorts and longer follow-up periods is necessary to validate these findings and to explore the therapeutic potential of targeting *TAL1* in ALL.

Conclusion

In summary, this study provides compelling evidence for the significant role of *TAL1* overexpression in the biogenesis of T-cell acute lymphoblastic leukemia (T-ALL) within the Iranian pediatric population. Furthermore, our findings highlight the potential utility of *TAL1* expression levels as a valuable biomarker for minimal residual disease (MRD) evaluation post-induction, offering a promising avenue for improved risk stratification and management of Iranian Pediatric T-ALL patients. Based on these results, we propose that *TAL1* expression analysis holds significant promise as a novel diagnostic and prognostic biomarker, warranting its consideration for integration into T-ALL treatment management strategies to potentially personalize therapy and enhance patient outcomes in this specific population.

Informed Consent

Written informed consent was obtained from all participants prior to their inclusion in the study.

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CONFLICT OF INTEREST: The authors declare no conflicts of interest related to this study. There are no financial, personal, or professional relationships that could have influenced the research design, execution, or interpretation. The study was conducted independently, without vested interests from funding sources, institutions, or individuals. The authors confirm they have not received compensation or benefits from entities that could gain from the publication of these findings. All data, analyses, and conclusions are based solely on scientific merit and objective research.

FINANCIAL SUPPORT: None

ETHICS STATEMENT: The current study was approved by the Research Ethics Committee, the Vice-Chancellor in Research Affairs at Shahid Beheshti University of Medical Sciences (Approval ID: IR.UT.SBMU.RETECH.REC.1402.627). The approval was granted on January 7, 2024.

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