Quantitative analysis of IKZF1 gene deletions in pediatric B-cell precursor acute lymphoblastic leukemia: Higher levels are associated with a poorer prognosis

Zhizhuo Huang (huang.cat@163.com)
Peking University People's Hospital https://orcid.org/0000-0003-4376-5399

Jia Yueping
Peking University People's Hospital

Ruan Guorui
Peking University People's Hospital

Zuo Yingxi
Peking University People's Hospital

Wu Jun
Peking University Shougang Hospital

Lu Aidong
Peking University People's Hospital

Xue Yujuan
Peking University People's Hospital

Cheng Yifei
Peking University People's Hospital

Le-ping Zhang
Peking University People's Hospital https://orcid.org/0000-0002-0754-9018

Research Article

Keywords: children, acute lymphoblastic leukemia, IKZF1 gene deletion, quantitative, clinical characteristics, prognosis

Posted Date: March 19th, 2021

DOI: https://doi.org/10.21203/rs.3.rs-321255/v1

Version of Record: A version of this preprint was published at Pediatric Hematology and Oncology on September 28th, 2021. See the published version at https://doi.org/10.1080/08880018.2021.1966558.

Abstract

Purpose: To assess the prognostic effect of different levels of IKZF1 gene deletions in pediatric B-cell precursor acute lymphoblastic leukemia (BCP-ALL).

Methods: IKZF1 Δ2-8/ALB deletions were quantified using multiplex real-time quantitative PCR in newly diagnosed pediatric BCP-ALL patients. Seventy-four patients with IKZF1 deletions ≥ 0.01% were included. Clinical characteristics, laboratory data, and treatment outcomes were analysed.

Results: The patients were divided into two groups: IKZF1 deletions < 1% (group A) and ≥ 1% (group B). Group B patients had a higher BCR-ABL1 positive rate than did group A patients. The proportions of patients who had an age at onset ≥10 years old, and white blood cell count ≥ 50×10^9/L were significantly higher in group B than in group A. The 3-year overall survival (OS) and 3-year event-free survival (EFS) rates in group B were 79 ± 8.8% and 62.4 ± 9.7%, respectively, being significantly lower than those in group A (97.7 ± 2.2% and 83.2 ± 5.8%, respectively). The level of IKZF1 deletions ≥ 1% and the central nervous system leukemia were independent risk factors of EFS.

Conclusions: Pediatric BCP-ALL patients with high levels of IKZF1 gene deletions have a poorer prognosis than those with low levels.

Introduction

Acute lymphoblastic leukemia (ALL) is the most common childhood malignant cancer, with B-cell precursor ALL (BCP-ALL) accounting for approximately 80% of pediatric ALL cases. The application of molecular biological and cytogenetic techniques has revealed that the translocations t(9;22)/(BCR-ABL1), t(12;21)/(ETV6/RUNX1), and t(1;19)/(E2A/PBX1); rearrangements of the MLL gene; and hypodiploid or hypodiploid karyotypes in certain patients with ALL are associated with different prognoses. Patients benefit from risk-adapted treatment protocols. Because of progress in chemotherapy regimens and the development of risk-adapted treatment protocols, the prognosis of ALL in children has significantly improved. The 5-year event-free survival (EFS) rate of pediatric ALL from developed countries can exceed 80% (Pui et al., 2014), but the remaining patients are still refractory/relapse with poor prognosis. Therefore, studies are currently focused on identifying high-risk ALL children in an early stage and carrying out the appropriate risk-adapted treatment to improve their prognosis.

The IKZF1 gene is located on chromosome 7p12 and encodes the lymphoid transcription protein Ikaros, which plays a key regulatory role in lymphopoiesis (Linda Olsson, 2015). Deletion or mutation of the IKZF1 gene is the hallmark of both BCR-ABL1-positive ALL and Ph-like ALL patients. In most cases, IKZF1 gene deletion has been recognised as a poor prognostic factor in pediatric ALL (Stanulla et al., 2020), Most data on IKZF1 deletions in pediatric ALL have been obtained through multiplex ligation probe-dependent amplification (MLPA) analyses, single nucleotide polymorphism (SNP) arrays, or polymerase chain reaction (PCR) assays, which cannot be used to detect minimal residual disease (MRD) in ALL. Recently, the levels of the IKZF1 Δ2–8/albumin (ALB) gene deletion have been detected using a real-time

quantitative polymerase chain reaction (RQ-PCR) and used to monitor MRD in ALL. In this study, a quantitative analysis of IKZF1 gene deletion was conducted in pediatric patients with BCP-ALL to explore the relationship between different levels of IKZF1 gene deletions and prognosis.

Materials And Methods

Patients

The study was approved by the Ethics Committee of Peking University People's Hospital, and the guardians of all patients provided written informed consent.
Seventy-four newly diagnosed pediatric BCP-ALL patients (aged < 18 years) with IKZF1 gene deletions, who underwent initial treatment in our institution between June 2014 and January 2018 were included in this study. All patients met the morphology-immunology-cytogenetics-molecular (MICM) criteria (Cui et al., 2018). The level of IKZF1 Δ2–8/ALB deletions in bone marrow (BM) samples was detected by multiplex real-time quantitative PCR (RQ-PCR), as described in a previous study.5 A patient with a IKZF1 deletion was defined as having IKZF1 Δ2–8/ALB deletions ≥ 0.01% of the BM sample at diagnosis. Clinical characteristics, laboratory data, and treatment outcomes were analysed. The study was approved by the Ethics Committee of Peking University People's Hospital, and the guardians of all patients provided written informed consent.

Treatment

Initially, all patients received the induction therapy referred to as improved ALL-Berlin-Frankfurt–Münster (BFM) protocol, as described earlier (Wang et al., 2018; Xue et al., 2019). Briefly, the patients received a CODPL induction (cyclophosphamide and prednisone or dexamethasone, vincristine, daunorubicin/idarubicin, and L-asparaginase) followed by consolidation and maintenance therapy. Two courses of 4-week re-induction therapy were administered every 6 months during consolidation treatment. The consolidation treatment consisted of 15 courses of high-dose methotrexate with or without pegaspargase, 3 courses of high-dose cytarabine, 2 courses of 4-week re-induction therapy, and a course of ifosfamide. Moreover, the patients received 22–25 rounds of methotrexate, cytarabine, and dexamethasone intrathecal therapy to prevent central nervous system leukemia (CNSL). The maintenance therapy consisted of oral 6-mercaptopurine daily and weekly intramuscular methotrexate administration. The whole treatment course lasted 3–3.5 years.

The patients who were BCR-ABL1-positive were stratified into the high risk (HR) group and others were stratified into the intermediate risk (IR) group when they began the induction therapy. Then risk stratification was further refined by MRD detection of BM samples at the 15th or 33rd day of induction chemotherapy (Winick and Devidas et al., 2017). IR patients with MRD ≥ 25% blast at the 15th day or MRD ≥ 1% blast at the 33rd day were upstaged to the HR group. The dose and intensity of anthracyclines and high-dose methotrexate differed by risk group.

Fourteen patients underwent allogeneic hematopoietic stem cell transplantation (allo-HSCT) after several courses of consolidation treatment. Ten patients received allo-HSCT after a first complete remission

(CR1), and four relapse patients chose allo-HSCT after achieving a second complete remission (CR2).

In addition to routine chemotherapy, the patients who were BCR-ABL1-positive also received a tyrosine kinase inhibitor (TKI) orally throughout the course, and some of the refractory/relapsed patients received TKIs irregularly. TKIs included the first-generation TKI imatinib mesylate (Novartis, Basel, Switzerland) (initial dose of 260–340 mg/m^2/d) and the second-generation TKI dasatinib (Bristol-Myers Squibb Company, Mount Vernon, USA) (initial dose of 50 mg/m^2/d).

MRD detection

MRD detection was performed by real-time quantitative PCR, and some common fusion transcripts, such as ETV6/RUNX1, E2A/PBX1, and BCR-ABL1, were assessed using reverse transcription PCR. Karyotyping screening was performed using a conventional karyotyping (G-banding) analysis or fluorescence in situ hybridisation (FISH). Monitoring of BM samples for MRD, common fusion transcripts, and IKZF1 Δ2–8/ALB deletions was carried out every 2–3 months during the first 3 years after diagnosis, after which these criteria were assessed every 6 months for 2 years.

Statistical analysis

Overall survival (OS) was defined as the time from diagnosis to the date of last follow-up or death from any cause. EFS was defined as the time from diagnosis to the first event (remission failure, relapse, death, or secondary malignancy) or the last follow-up. Continuous variables were expressed as medians and ranges, while categorical variables were expressed as frequencies and percentages. The differences between different groups were analysed using a Mann-Whitney U-test or a Chi-square test for continuous and categorical variables, respectively. OS and EFS were analysed using the Kaplan-Meier method and a log-rank test. Cox regression analysis was performed using a multivariate analysis. Statistical analysis was performed using SPSS software (version 22.0; IBM, Armonk, NY). A P value of < 0.05 was considered statistically significant.

Results

Patient characteristics

The median age of the 74 patients (46 boys and 28 girls) with IKZF1 Δ2–8/ALB deletions was 8 years (range, 1–17 years). The last follow-up was July 1, 2020, and the median follow-up period was 38 months (range, 2–72 months). The clinical and laboratory characteristics of the patients are summarised in Table 1.

Table 1
Clinical, laboratory characteristics of patients with different levels of IKZF1 Δ2–8/ALB deletions

Variant	Total (n = 74)	IKZF1/ALB<1% (n = 45)	IKZF1/ALB ≥ 1% (n = 29)	*P*
Male (%)	46(62.2%)	25(55.6%)	21(72.4%)	0.144
Female (%)	28(37.8%)	20(44.4%)	8(27.6%)	
Median age (years)(range)	8(1–17)	6(1–16)	11(1–17)	0.031
1 to 10 yeas (%)	41(55.4%)	28(62.2%)	13(44.8%)	0.142
≥10 yeas (%)	33(44.6%)	17(37.8%)	16(55.2%)	
WBC count (×10^9/L) (%)	58 (78.4%)	39(86.7%)	19(65.5%)	0.031
<50	16(21.6%)	6(13.3%)	10(34.5%)	
≥50				
LDH (U/L)	545.0 ± 479.5	515.8 ± 437.7	519.0 ± 544.0	0.520
UA (umol/L)	370.3 ± 138.6	348.7 ± 108.2	404.2 ± 173.0	0.137
Immunologic subtype (%)	68(91.9%)	41(91.1%)	27(93.1%)	0.405
Com-B-ALL	3(4.05%)	1(2.2%)	2(6.9%)	
Pre-B-ALL	3(4.05%)	3(6.7%)	0(0%)	
Pro-B-ALL				
Fusion transcripts (%)	10(13.5%)	1(2.2%)	9(31.0%)	0.01
BCR-ABL1	1(1.4%)	1(2.2%)	0(0%)	
E2A/PBX1	8(10.8%)	8(17.8%)	0(0%)	
ETV6/RUNX1	55(74.3%)	35(77.8%)	20(69.0%)	
Other				
Karyotype (%)	11(14.9%)	8(17.8%)	3(10.3%)	0.501
Hyperdiploid	11(14.9%)	8(17.8%)	3(10.3%)	
Hypodiploid	29(39.2%)	15(33.3%)	14(48.3%)	
Normal	23(31.0%)	14(31.1%)	9(31.1%)	
Other				

Abbreviations: LDH, lactate dehydrogenase. UA, uricacid. CNSL, central nervous system leukemia. TKI, tyrosine kinase inhibitor. HSCT, hematopoietic stem cell transplantation.

Variant	Total (n = 74)	IKZF1/ALB＜1% (n = 45)	IKZF1/ALB ≥ 1% (n = 29)	*P*
MRD on 15 days (%)	26(35.1%)	15(33.3%)	11(37.9%)	0.686
MRD ≥ 5%	48(64.9%)	30(66.7%)	18(62.1%)	
MRD < 5%				
MRD on 33 days (%)	43(58.1%)	27(60.0%)	16(55.2%)	0.797
MRD ＜0.01%	23(31.1%)	14(31.1%)	9(31.0%)	
0.01% ≤MRD ＜1%	8(10.8%)	4(8.9%)	4(13.8%)	
MRD ≥ 1%				
Risk stratification (%)	53(71.6%)	39 (86.7%)	14(48.3%)	0.001
IR	21(28.4%)	6(13.3%)	15(51.7%)	
HR				
CNSL (%)	14(18.9%)	6(13.3%)	8(27.6%)	0.126
Yes	60(81.1%)	39(86.7%)	21(72.4%)	
No				
TKI (%)	27(36.5%)	7(15.6%)	20(69.0%)	< 0.001
Yes	47(63.5%)	38(84.4%)	9(31.0%)	
No				
HSCT (%)	14(18.9%)	5(11.1%)	9(31.0%)	0.033
Yes	60(81.1%)	40(88.9%)	20(69.0%)	
No				
Relapse/Refractory (%)	16(21.6%)	6(13.3%)	10(34.5%)	0.031
Yes	58(78.4%)	39(86.7%)	19(65.5%)	
No				
Death (%)	6(8.1%)	1(2.2%)	5(17.2%)	0.020
Yes	68(91.9%)	44(97.8%)	24(82.8%)	
No				

Abbreviations: LDH, lactate dehydrogenase. UA, uricacid. CNSL, central nervous system leukemia. TKI, tyrosine kinase inhibitor. HSCT, hematopoietic stem cell transplantation.

The distribution of IKZF1 Δ2–8/ALB gene deletion levels

The distribution of IKZF1 deletion levels is shown in Table 2. The level of IKZF1 deletions in the 74 patients was 0.01–243.76%. There were 45 patients (60.8%) who had IKZF1Δ2–8/ALB deletions < 1%, including only a single patient (2.2%) who was BCR-ABL1 fusion gene-positive, while the others were negative. Of the 29 patients with IKZF1Δ2–8/ALB deletions ≥ 1%, nine patients (31.0%) were BCR-ABL1 fusion gene-positive. Patients with IKZF1Δ2–8/ALB deletions ≥ 1% had a higher BCR-ABL1 positive rate than those with IKZF1Δ2–8/ALB deletions < 1% (31.0% vs. 2.2%, $P = 0.001$), as shown in Fig. 1.

Table 2
The distribution for different levels of IKZF1 deletions in 74 patients

IKZF1/ALB, %	Total, n (%)
0.01–0.1	33(44.6%)
0.1-1	12(16.2%)
1–10	5(6.8%)
≥ 10	24(32.4%)

IKZF1 Δ2–8/ALB gene deletions and clinical features

The patients were divided into two groups: IKZF1 Δ2–8/ALB deletions < 1% (group A) and ≥ 1% (group B). The rates of age at onset ≥ 10 years old, white blood cell count (WBC) ≥ 50×10^9/L at initial diagnosis, and HR patients in group B were significantly higher than those in group A ($P < 0.05$). Patients who were positive for the ETV6/RUNX1 fusion gene in group A accounted for 17.8%, which was higher than that in group B. However, the level of IKZF1 deletions was not associated with sex, WBC count, lactate dehydrogenase (LDH), uric acid (UA), immunologic subtype, or karyotype. The CNSL rate between the two groups were similar. More patients were treated with TKIs and underwent HSCT in group B than in group A ($P < 0.05$). The characteristics of patients with BCP-ALL with different IKZF1 Δ2–8/ALB deletion levels are summarised in Table 1.

IKZF1 Δ2–8/ALB gene deletions and MRD

There were no statistically significant differences between the two groups in the level of MRD at the 15th or 33rd day of induction chemotherapy or in the incidence of CNSL. In group B, 34.5% of the patients were refractory/relapsed, which was significantly higher than that in group A (34.5% vs. 13.3%, $P = 0.031$). The mortality in group B was significantly higher than that in group A (17.2% vs. 2.2%, $P = 0.02$), as shown in Table 1.

OS, EFS, and IKZF1 Δ2–8/ALB gene deletions

The 3-year OS and 3-year EFS in Group B were 79.3 ± 8.8% and 62.4 ± 9.7%, respectively, and these were significantly lower than the 3-year OS (97.7 ± 2.2%, $P = 0.022$) and 3-year EFS (83.2 ± 5.8%, $P = 0.019$) in

group A (Fig. 2). The 3-year OS and 3-year EFS of Ph-negative patients in group B were also lower than the 3-year OS (97.6 ± 2.2% vs. 84.7 ± 8.1%, $P = 0.014$) and 3-year EFS (85.2 ± 5.6% vs. 59.0 ± 12.9%, $P = 0.013$) of Ph-negative patients in group A (Fig. 3). Multivariate analysis revealed that IKZF1 deletions ≥ 1% and CNSL were independent risk factors of EFS, while factors such as age, gender, WBC count, CNSL, BCR-ABL1 fusion gene, risk stratification, and the level of IKZF1 deletions were not associated with OS (Table 3).

Table 3
Multivariate analysis of possible prognostic factors of OS and EFS

Factors	OS			EFS		
	HR	95% CI	*P*	HR	95% CI	*P*
Male	0.739	0.066–8.317	0.807	0.627	0.188–2.092	0.448
Age ≥ 10 years	1.907	0.284–12.795	0.506	2.087	0.753–5.785	0.157
WBC ≥ 50×10^9/L	4.136	0.424–40.387	0.222	1.277	0.359–4.537	0.706
BCR-ABL1 $^+$	0.046	0.002–1.354	0.074	0.668	0.100-4.468	0.678
MRD ≥ 1% on 33 days	1.927	0.222–16.710	0.552	1.142	0.164–7.963	0.894
High-risk	1.480	0.129–16.924	0.753	0.742	0.149–3.702	0.716
IKZF1 deletions of ≥ 1%	8.842	0.922–84.806	0.059	3.054	1.015–9.188	0.047
CNSL	4.720	0.417–53.425	0.210	3.588	1.129–11.399	0.030
HSCT	2.015	0.206–19.728	0.547	0.659	0.169–2.569	0.547

Discussion

IKZF1 is composed of eight exons and encodes the transcription factor Ikaros (Payne, 2011). IKZF1 alterations can occur frequently in germline cells and somatic cells, and it has been studied extensively. IKZF1 alterations in somatic cells are found in approximately 15% of pediatric patients with ALL (Mullighan et al., 2009; Iacobucci et al., 2017), but the alternations increase in prevalence among adult patients with ALL. The most common types of IKZF1 alterations are referred to as focal deletions, such as the whole gene and exons 4–7 deletions, resulting in loss-of-function in Ikaros, whereas IKZF1 mutations are less reported, including missense mutations, nonsense mutations, and frameshift mutations (Linda Olsson, 2015). IKZF1 deletions are important molecular genetic makers in the development and relapse of ALL and attracted significant attention until Mullighan et al. first described such deletions in patients with BCR-ABL1-positive ALL in 2008 (Mullighan et al., 2008). Overall, the prevalence of IKZF1 deletions is 10–30% in pediatric patients with B-ALL who are BCR-ABL1-negative (Mullighan et al., 2009), and can be as high as 56.8–70.6% and 27–68% in pediatric patients with B-ALL

who are BCR-ABL1-positive and who have Ph-like ALL, respectively. These patients are often associated with high WBC counts, older age, poor chemotherapy responses, higher risks of relapse, and insensitivity to TKIs (Mullighan, 2012; van der Veer et al., 2014). In contrast, among ALL subtypes with good prognosis, the prevalence of IKZF1 deletions is low, accounting for 3–6% and 15% of ETV6-RUNX1 and hyperdiploid cases, respectively (Mullighan et al., 2009; Dorge et al., 2013; Enshaei et al., 2013). Thus, for a long time, IKZF1 gene deletions have been established to be an independent adverse prognostic factor for patients with ALL (Stanulla et al., 2020; Vairy et al., 2020). Therefore, screening for IKZF1 deletions is very important in recognising high-risk patients and initiating risk-adapted chemotherapy for pediatric patients with ALL.

Most of the published data on IKZF1 deletions have been generated using MLPA analyses, SNP arrays, or multiplex PCR assays. Previous results have shown that RQ-PCR based on ALB as a control gene is a reliable and sensitive method for detecting IKZF1 deletions (Li-Xin Wu, 2017), and there is a good correlation between quantification of MRD and the level of IKZF1 deletions (Venn et al., 2012; Caye et al., 2013; Li-Xin Wu, 2017). In this study, we conducted a quantitative analysis of IKZF1 deletions in pediatric patients with BCP-ALL to assess the impact of different levels of IKZF1 deletions on prognosis. The level of IKZF1 deletions in our patients ranged from 0.01 to 243.76%, with 60.8% of the patients being in the IKZF1 deletions < 1% group. The rates of IKZF1 deletions in patients with ALL who were ETV6-RUNX1-positive or hyperdiploid were 10.8% and 14.9%, respectively, which is consistent with previous observations. Overall, 13.5% of the patients were BCR-ABL1-positive, but patients with IKZF1 deletions ≥ 1% had a significantly higher BCR-ABL1-positive rate (31.0%) than those with IKZF1 deletions < 1% (2.2%). In contrast, all patients who were ETV6/RUNX1-positive had IKZF1 deletions < 1%. Wu et al (Li-Xin Wu, 2017). reported that 90% of adult patients with IKZF1 deletions had levels of IKZF1 deletions ≥ 1%, and the rate of IKZF1 deletions in patients who were BCR-ABL1-positive was 68.8%. These results differ from ours. One reason for this may be that patients who were in the high-risk group and were BCR-ABL1-positive were more likely to have high levels of IKZF1 deletions; therefore, in our study, fewer BCR-ABL1-positive pediatric patients with ALL were in the high-risk group than did adult patients.

In our study, the percentages of patients whose age at onset was ≥ 10 years old, had WBC ≥ 50×10^9/L at initial diagnosis, and were in high-risk group were significantly higher in the IKZF1 deletions ≥ 1% group than in the IKZF1 deletions < 1% group. In contrast, there were no differences in other clinical and laboratory characteristics between the two groups. Various clinical trials have shown that IKZF1 deletions are associated with an older age at diagnosis, a higher WBC count, higher levels of MRD after induction, and a higher risk of relapse, which corresponded with our study (Dorge et al., 2013; van der Veer et al., 2013; Olsson et al., 2014; van der Veer et al., 2014; Linda Olsson, 2015; Olsson et al., 2015). We found no difference in MRD at 15 and 33 days between the two groups of patients with different levels of IKZF1 deletions, whereas the rate of relapse/refractoriness in the IKZF1 deletions ≥ 1% group was significantly higher than that in the IKZF1 deletions < 1% group, suggesting that there was more HR ALL in the IKZF1 deletions ≥ 1% group.

Almost all clinical studies have demonstrated that IKZF1 deletions are an independent adverse prognostic factor, especially in combination with an early MRD response, in pediatric patients with BCP-ALL who are either BCR-ABL1-positive or negative. In children with BCR-ABL1-negative ALL who were treated using the BFM-2000 protocol (Dorge et al., 2013), the 5-year EFS was 69% for patients with IKZF1 alterations vs. 85% for patients without IKZF1 alterations. Patients with IKZF1 alterations also had a higher incidence of relapse (21% vs. 10%). Similar poor outcomes in pediatric patients with IKZF1 deletions were observed in a Japanese pediatric Ph-negative B-ALL cohort (Asai et al., 2013), the AEIOP-BFM 2000 cohort (Palmi et al., 2013), and the IC-BFM-2002 cohort. Of note, the ETV6-RUNX1 subtype should be ruled out in patients with Ph-negative B-ALL who also have IKZF1 alterations because they have favourable outcomes despite the presence of IKZF1 alterations (Lilljebjorn et al., 2016). Therefore, the prognosis is still poor for BCR-ABL1–positive and Ph-like patients who have IKZF1 deletions, even if they are treated with TKIs. In AALL0622, a Children's Oncology Group (COG) phase II trial, patients with ALL who were Ph + and also had IKZF1 deletions were treated with dasatinib plus intensive chemotherapy and were found to have inferior 5-year EFS and OS compared to those patients with ALL who were Ph + but had wild type IKZF1 (52% vs. 82%, $P = 0.04$ and 80% vs. 100%, $P = 0.04$, respectively) (Slayton et al., 2018). Our results also showed that the 3-year OS and 3-year EFS in the IKZF1 deletions ≥ 1% group were both significantly poorer than the 3-year OS (79.3% vs. 97.7%, $P = 0.022$) and 3-year EFS (62.4% vs. 83.2%, $P = 0.019$) in the IKZF1 deletions < 1% group. Similar results were found in BCR-ABL1-negative patients with corresponding levels of IKZF1 deletions. Therefore, the prognosis is worse in patients with a higher percentage of IKZF1 deletions, regardless of BCR-ABL1 positivity or negativity. Because there was only one BCR-ABL1-positive patient in the IKZF1 deletions < 1% group, we could not compare the effect of different levels of IKZF1 deletions on the prognosis of BCR-ABL1-positive children. Multivariate analysis showed that the level of IKZF1 deletions ≥ 1% and CNSL were independent risk factors of EFS. The level of IKZF1 deletions ≥ 1% is likely to be a poor prognosis factor of OS, although no significant statistical difference was indicated due to a limited sample size. In the future, studies on the effect of different levels of IKZF1 deletions on prognosis with large cohorts of pediatric patients with BCP-ALL should be carried out. In addition, pediatric BCP-ALL patients with the level of IKZF1 deletions ≥ 1% may be stratified into high-risk groups due to poor prognosis to receive intensive chemotherapy.

Our study has several limitations. First, this was a single-centre study with a small sample size. Second, due to the limitation of sample size, comparison between subgroups could not be performed.

In conclusion, pediatric BCP-ALL patients with IKZF1 Δ2–8/ALB deletions < 1% had a good prognosis, whereas patients with IKZF1 Δ2–8/ALB deletions ≥ 1% had a poor outcome, and these patients always had other risks factors, such as older age, higher WBC count, positivity for the BCR/ABL1 fusion gene, and a higher relapse rate.

Declarations

Conflicts of interest: The authors have no conflicts of interest to declare.

References

Asai, D. and T. Imamura, et al. (2013). "IKZF1 deletion is associated with a poor outcome in pediatric B-cell precursor acute lymphoblastic leukemia in Japan." Cancer Med 2 (3): 412-9.

Caye, A. and K. Beldjord, et al. (2013). "Breakpoint-specific multiplex polymerase chain reaction allows the detection of IKZF1 intragenic deletions and minimal residual disease monitoring in B-cell precursor acute lymphoblastic leukemia." Haematologica 98 (4): 597-601.

Cui, L. and Z. G. Li, et al. (2018). "Outcome of children with newly diagnosed acute lymphoblastic leukemia treated with CCLG-ALL 2008: The first nation-wide prospective multicenter study in China." Am J Hematol 93 (7): 913-920.

Dorge, P. and B. Meissner, et al. (2013). "IKZF1 deletion is an independent predictor of outcome in pediatric acute lymphoblastic leukemia treated according to the ALL-BFM 2000 protocol." Haematologica 98 (3): 428-32.

Enshaei, A. and C. J. Schwab, et al. (2013). "Long-term follow-up of ETV6-RUNX1 ALL reveals that NCI risk, rather than secondary genetic abnormalities, is the key risk factor." Leukemia 27 (11): 2256-9.

Iacobucci, I. and C. G. Mullighan (2017). "Genetic Basis of Acute Lymphoblastic Leukemia." J Clin Oncol 35 (9): 975-983.

Lilljebjorn, H. and R. Henningsson, et al. (2016). "Identification of ETV6-RUNX1-like and DUX4-rearranged subtypes in paediatric B-cell precursor acute lymphoblastic leukaemia." Nat Commun 7: 11790.

Linda Olsson, B. J. (2015). "Ikaros and leukaemia." Br J Haematol 169 (4): 479-491.

Li-Xin Wu, J. Z. Q. Y. (2017). "High frequency of IKZF1 deletions in Chinese adult patients with acute lymphoblastic leukemia detected by multiplex realtime quantitative PCR." Clinical and Diagnostic Pathology 1 (2): 1-7.

Mullighan, C. G. (2012). "The molecular genetic makeup of acute lymphoblastic leukemia." Hematology Am Soc Hematol Educ Program 2012: 389-96.

Mullighan, C. G. and C. B. Miller, et al. (2008). "BCR-ABL1 lymphoblastic leukaemia is characterized by the deletion of Ikaros." Nature 453 (7191): 110-4.

Mullighan, C. G. and X. Su, et al. (2009). "Deletion of IKZF1 and prognosis in acute lymphoblastic leukemia." N Engl J Med 360 (5): 470-80.

Olsson, L. and A. Castor, et al. (2014). "Deletions of IKZF1 and SPRED1 are associated with poor prognosis in a population-based series of pediatric B-cell precursor acute lymphoblastic leukemia diagnosed between 1992 and 2011." Leukemia 28 (2): 302-10.

Olsson, L. and O. I. Ivanov, et al. (2015). "The clinical impact of IKZF1 deletions in paediatric B-cell precursor acute lymphoblastic leukaemia is independent of minimal residual disease stratification in Nordic Society for Paediatric Haematology and Oncology treatment protocols used between 1992 and 2013." Br J Haematol 170 (6): 847-58.

Palmi, C. and M. G. Valsecchi, et al. (2013). "What is the relevance of Ikaros gene deletions as a prognostic marker in pediatric Philadelphia-negative B-cell precursor acute lymphoblastic leukemia?" Haematologica 98 (8): 1226-31.

Payne, M. A. (2011). "Zinc finger structure-function in Ikaros Marvin A Payne." World J Biol Chem 2 (6): 161-6.

Pui, C. H. and D. Pei, et al. (2014). "A revised definition for cure of childhood acute lymphoblastic leukemia." Leukemia 28 (12): 2336-43.

Slayton, W. B. and K. R. Schultz, et al. (2018). "Dasatinib Plus Intensive Chemotherapy in Children, Adolescents, and Young Adults With Philadelphia Chromosome-Positive Acute Lymphoblastic Leukemia: Results of Children's Oncology Group Trial AALL0622." J Clin Oncol 36 (22): 2306-2314.

Stanulla, M. and H. Cave, et al. (2020). "IKZF1 deletions in pediatric acute lymphoblastic leukemia: still a poor prognostic marker?" Blood 135 (4): 252-260.

Vairy, S. and T. H. Tran (2020). "IKZF1 alterations in acute lymphoblastic leukemia: The good, the bad and the ugly." Blood Rev 44: 100677.

van der Veer, A. and E. Waanders, et al. (2013). "Independent prognostic value of BCR-ABL1-like signature and IKZF1 deletion, but not high CRLF2 expression, in children with B-cell precursor ALL." Blood 122 (15): 2622-9.

van der Veer, A. and M. Zaliova, et al. (2014). "IKZF1 status as a prognostic feature in BCR-ABL1-positive childhood ALL." Blood 123 (11): 1691-8.

Venn, N. C. and V. H. van der Velden, et al. (2012). "Highly sensitive MRD tests for ALL based on the IKZF1 Delta3-6 microdeletion." Leukemia 26 (6): 1414-6.

Wang, Y. and H. M. Zeng, et al. (2018). "ETV6/RUNX1-positive childhood acute lymphoblastic leukemia in China: excellent prognosis with improved BFM protocol." Ital J Pediatr 44 (1): 94.

Winick, N. and M. Devidas, et al. (2017). "Impact of Initial CSF Findings on Outcome Among Patients With National Cancer Institute Standard- and High-Risk B-Cell Acute Lymphoblastic Leukemia: A Report From the Children's Oncology Group." J Clin Oncol 35 (22): 2527-2534.

Xue, Y. J. and Y. F. Cheng, et al. (2019). "Allogeneic Hematopoietic Stem Cell Transplantation, Especially Haploidentical, May Improve Long-Term Survival for High-Risk Pediatric Patients with Philadelphia

Chromosome-Positive Acute Lymphoblastic Leukemia in the Tyrosine Kinase Inhibitor Era." Biol Blood Marrow Transplant 25 (8): 1611-1620.

Figures

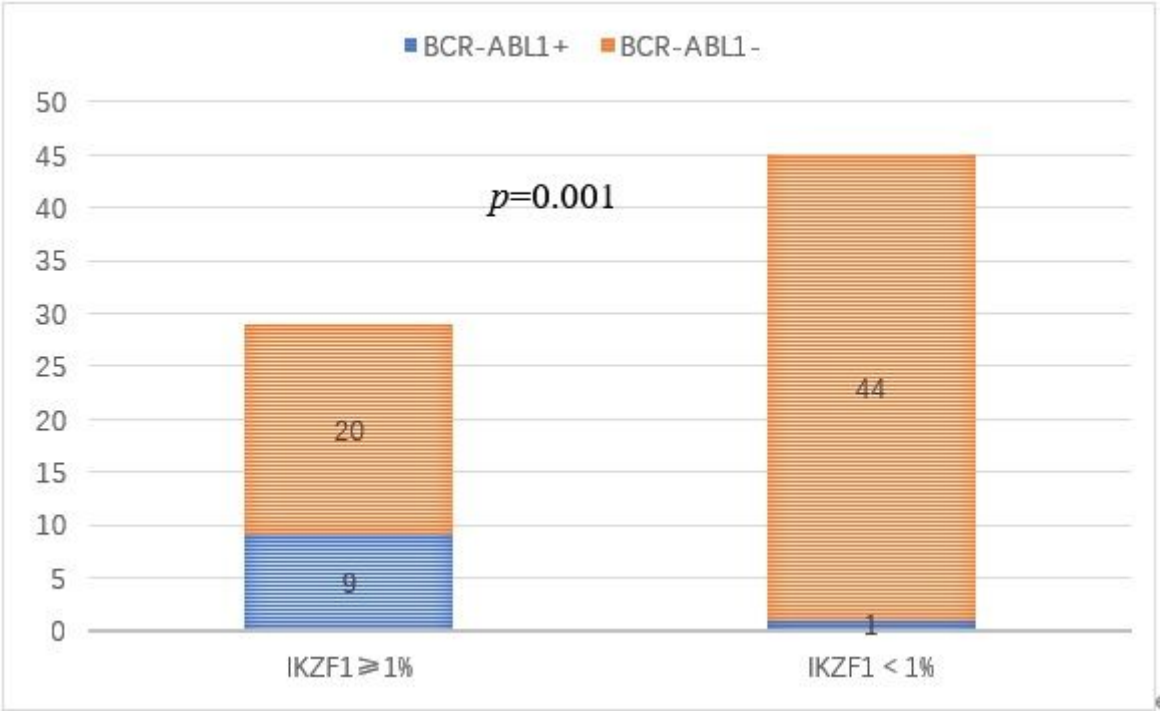

Figure 1

The distribution of different levels of IKZF1 deletions in patients with BCR-ABL1 positivity and negativity.

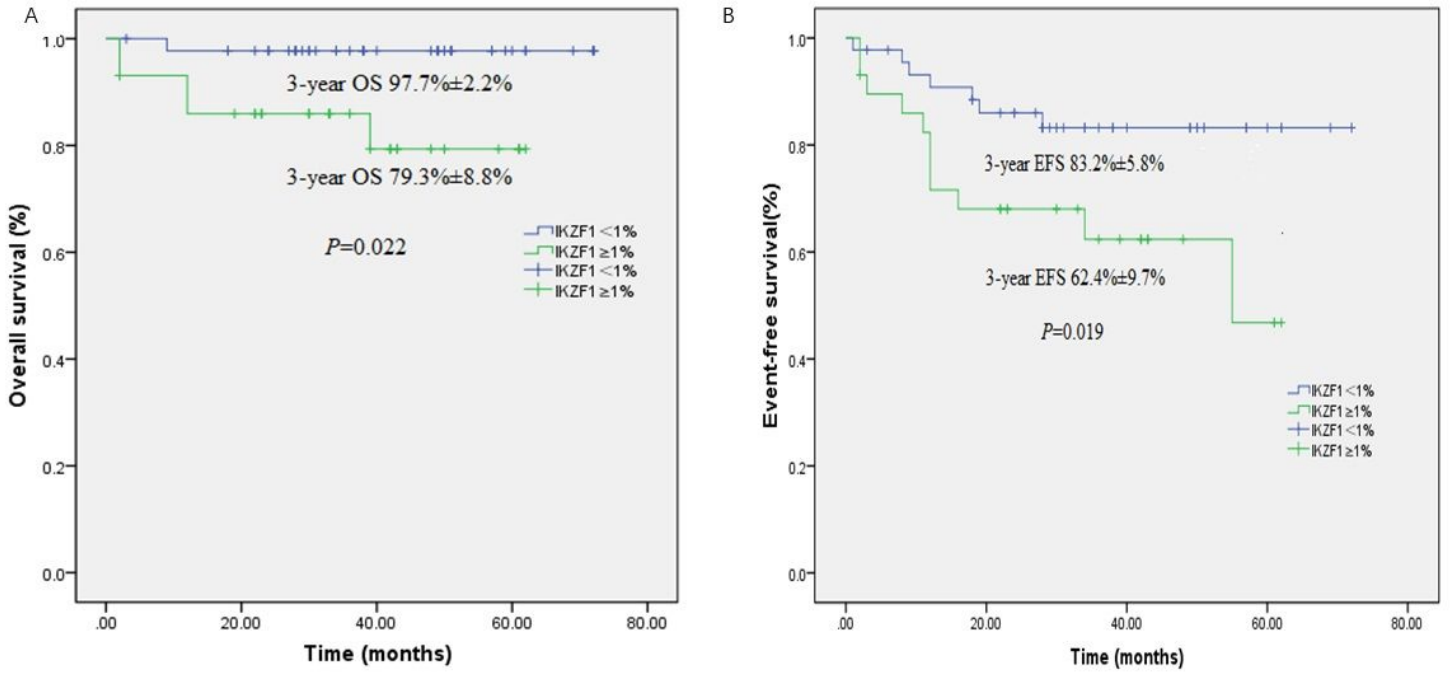

Figure 2

Kaplan-Meier survival curve of OS (a) and EFS (b) of the patients in the IKZF1 deletions ≥1% group and IKZF1 deletions＜1% group.

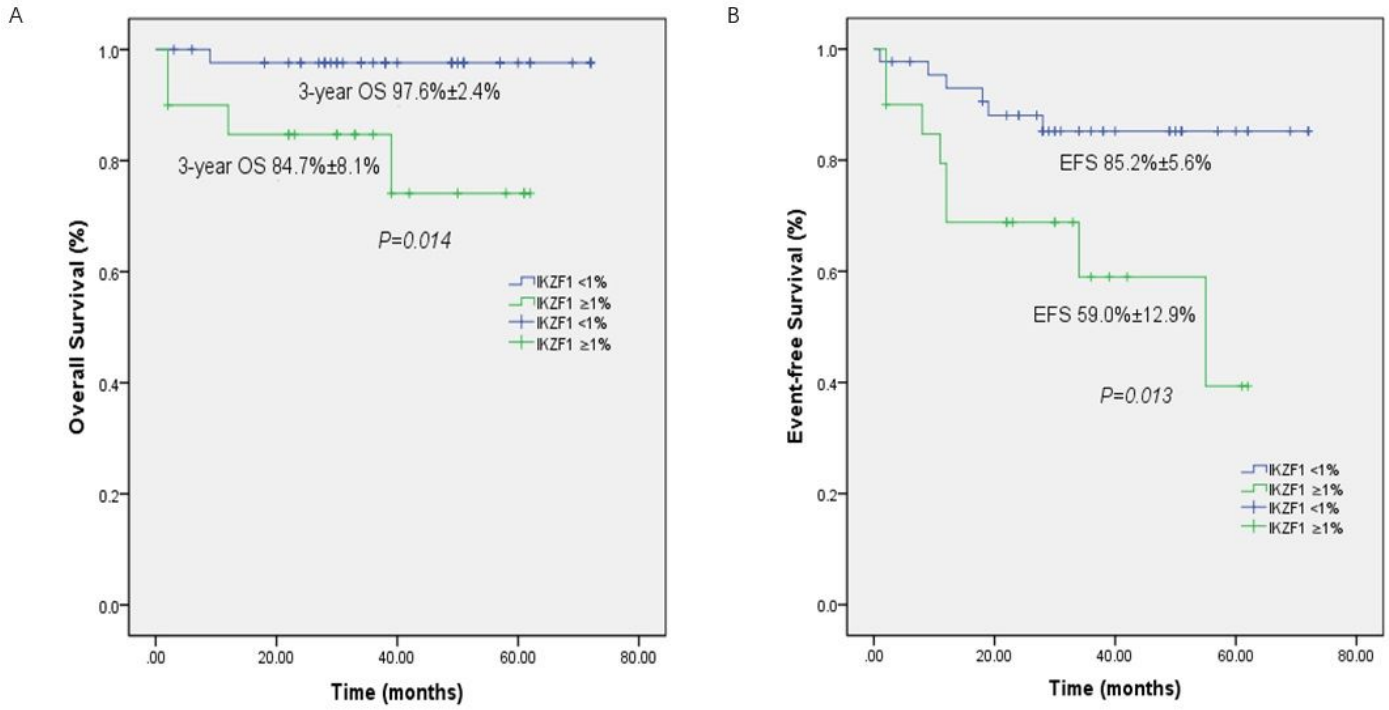

Figure 3

Kaplan-Meier survival curve of OS (a) and EFS (b) of BCR-ABL1-negative patients in the IKZF1 deletions ≥1% group and IKZF1 deletions＜1% group.