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# New precision medicine weapons for targeted treatment of high-risk B-cell precursor acute lymphoblastic leukemia

*by Sinisa Dovat and Joseph Schramm*

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**New precision medicine weapons for targeted treatment of high-risk B-cell precursor acute lymphoblastic leukemia**

Sinisa Dovat and Joseph Schramm

Penn State University College of Medicine, Hershey, PA

**Corresponding author**
Sinisa Dovat - sdovat@pennstatehealth.psu.edu

**Authors' contribution**

Authors equally contributed

**Disclosures**

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In this issue of *Haematologica*, Vervoort *et al* [1]. demonstrate that deletion of IKZF1 (IKZF1-del) in B-cell precursor acute lymphoblastic leukemia (BCP-ALL) promotes resistance to AraC. They further show that inhibition of either CDK8/19 kinases or casein kinase 2 restores sensitivity of IKZF1-del BCP-ALL to AraC treatment. BCP-ALL is the most common childhood malignancy. Despite improved outcomes, more children die from BCP-ALL than from any other type of malignancy. In adults, BCP-ALL is an aggressive disease with high mortality and requires highly toxic treatment. Advances in targeted chemotherapy treatment along with immunotherapy, greatly improved the overall prognosis of BCP-ALL in both children and adults [2,3]. However, relapsed BCP-ALL remains a clinical challenge, and is associated with high mortality [4]**.** IKZF1 encodes IKAROS, a master regulator of hematopoiesis and a tumor suppressor protein [5,6]. Deletion of a single copy of IKZF1 (IKZF1-del) results in high-risk BCP-ALL, which very often has a gene expression profile that is characteristic of Ph-like BCP-ALL which is associated with a high relapse rate and a poor outcome [7]**.** Chemotherapy resistance has been observed in IKZF1-del BCP-ALL and it is a common feature of this subtype of BCP-ALL [8]**.** Several mechanisms have been proposed to explain those observations, along with the therapeutic strategies to overcome resistance to particular chemotherapeutics [9].

To understand the mechanism of chemoresistance in BCP-ALL due to IKZF1 deletion, Vervoort et al. used genetic (by CRISPR/Cas9) and a pharmacological approach (using Iberdomid) to target IKAROS. They tested IKAROS-null BCP-ALL cells for sensitivity to common chemotherapeutics. They showed that IKZF1 absence is strongly associated with increased resistance to AraC. The causative role of IKZF1 deletion in increased resistance to AraC was demonstrated both in vitro and in vivo using patient-derived xenografts (PDXs) and a retrospective analysis of minimal residual disease (MRD) in patients. Using a functional approach, the authors identified two mechanisms through which loss of Ikaros drives resistance to AraC treatment (Fig. 1): 1) Decreased incorporation of cytarabine (AraC) due to lower expression of the solute carrier hENT1. The expression of this transporter is critical for AraC influx, at the doses of AraC used for the treatment of BCP-ALL. The authors found that IKZF1-del cells have reduced AraCTP incorporation, which can explain the mechanism of resistance to AraC in those cells. Reduced expression of hENT1 has been associated with AraC sensitivity in both acute myeloid leukemia (AML) and BCP-ALL; 2) Increased expression of the Evi1 oncogene. Elevated expression of Evi1 has been associated with resistance to AraC and poor prognosis in AML. In addition, Evi1 functions as a regulator of hematopoietic stem cell self-renewal. Although the function of Evi1 has been studied extensively in AML and AML stem cells, alterations in Evi1 have not been associated with BCP-ALL. Based on presented data, it is likely that Evi1 repression is also important for normal B cell differentiation and that increased Evi1 expression can promote stemness and drug resistance in BCP-ALL. Since IKZF1 is located at chromosome 7p, and IKZF1 mutations have been observed in pediatric AML, these data suggest a potential mechanism of IKAROS tumor suppressor activity in AML – via repression of Evi1 expression.

The authors search for a way to restore sensitivity to AraC in IKZF1-null BCP-ALL (Fig. 1). They used a CRISPR/Cas9-based loss-of-function screen that targets all human kinases. They identified that the loss of homologous cyclin-dependent kinases CDK8/19 restores sensitivity of IKZF1-null BCP-ALL to AraC. The use of small molecule inhibitor CCT251921, which specifically targets CDK8 and 19, in combination with AraC, showed a strong synergistic effect in both IKZF1-wt and IKZF1-null BCP-ALL cells. This suggests that this novel combination can be a potent treatment for different types of high-risk BCP-ALL, regardless of *IKZF1* status. Additional findings showed that loss of casein kinase II (CK2), restores sensitivity to AraC in *IKZF1*-null BCP-ALL. Combination treatment with CK2 inhibitor Silmitasertib and AraC showed strong synergistic cytotoxicity in both IKZF1-wt and IKZF1-null BCP-ALL. It has been shown previously, that CK2 inhibition can sensitize IKZF1 haploinsufficient BCP-ALL via enhancement of IKAROS protein function produced from the remaining allele [10]. The data presented in this

paper suggest that CK2 inhibition can induce chemosensitivity of BCP-ALL cells via other mechanisms. It is intriguing that CK2 enhances Evi1 activity through direct phosphorylation, which could be an additional mechanism of CK2 inhibition in leukemia.

This paper has both basic/translational and clinical impact on the BCP-ALL field in many different ways: 1) Identifies a novel mechanism of drug resistance to chemotherapy of high-risk BCP-ALL. As the authors mentioned in the Discussion – awareness that IKZF1-del BCP-ALL is resistant to AraC treatment due to hENT1 repression suggests utilization of differing nucleoside analogues (ie fludarabine and clofarabine) will be beneficial for patients since they are imported by other transporters; 2) A potential novel targeted treatment for BCP-ALL. The data provide a rationale for preclinical testing of Silmitastetib and AraC combination therapy for BCP-ALL. Silmitasertib has been tested in Phase I/II trials for other malignancies, which could further accelerate testing of this combination treatment in clinical trials; 3) The role of Evi1 in BCL-ALL pathogenesis. The results suggest a potential role of Evi1 in BCP-ALL drug resistance. Further testing of the utility of Evi1 expression as a biomarker for drug-resistant BCP-ALL is warranted, and 4) The role of IKZF1/IKAROS in AML pathogenesis. Most of the research concerning the tumor suppression role of IKAROS is focused on BCP-ALL, despite the evidence for the role of IKZF1 in the pathogenesis of AML. The presented data suggest that IKZF1 can regulate expression of Evi1, a known oncogene in AML, which provides further support for studying the role of IKZF1 in AML. Overall, the studies by Vervoort *et al.* allow a better understanding of the role of IKZF1 in chemoresistance of BCP-ALL and also provide a rationale for preclinical and clinical testing of these novel combination treatments for this disease.

References


1 Vervoort BMT, Butler M, Grünewald KJT, et al. IKZF1 gene deletions drive resistance to cytarabine in B-cell precursor acute lymphoblastic leukemia. Haematologica. xxx

2 Pieters R, Mullighan CG, Hunger SP. Advancing Diagnostics and Therapy to Reach Universal Cure in Childhood ALL. J Clin Oncol. 2023;41(36):5579-5591.

3 Gokbuget N, Boissel N, Chiaretti S, et al. Diagnosis, prognostic factors, and assessment of ALL in adults: 2024 ELN recommendations from a European expert panel. Blood. 2024;143(19):1891-1902.

4 Wudhikarn K, King AC, Geyer MB, et al. Outcomes of relapsed B-cell acute lymphoblastic leukemia after sequential treatment with blinatumomab and inotuzumab. Blood Adv. 2022;6(5):1432-1443.

5 Georgopoulos K, Moore DD, Derfler B. Ikaros, an early lymphoid-specific transcription factor and a putative mediator for T cell commitment. Science. 1992;258(5083):808-812.

6 Lo K, Landau NR, Smale ST. LyF-1, a transcriptional regulator that interacts with a novel class of promoters for lymphocyte-specific genes. Mol Cell Biol. 1991;11(10):5229-5243.

7 Paietta E, Roberts KG, Wang V, et al. Molecular classification improves risk assessment in adult BCR-ABL1-negative B-ALL. Blood. 2021;138(11):948-958.

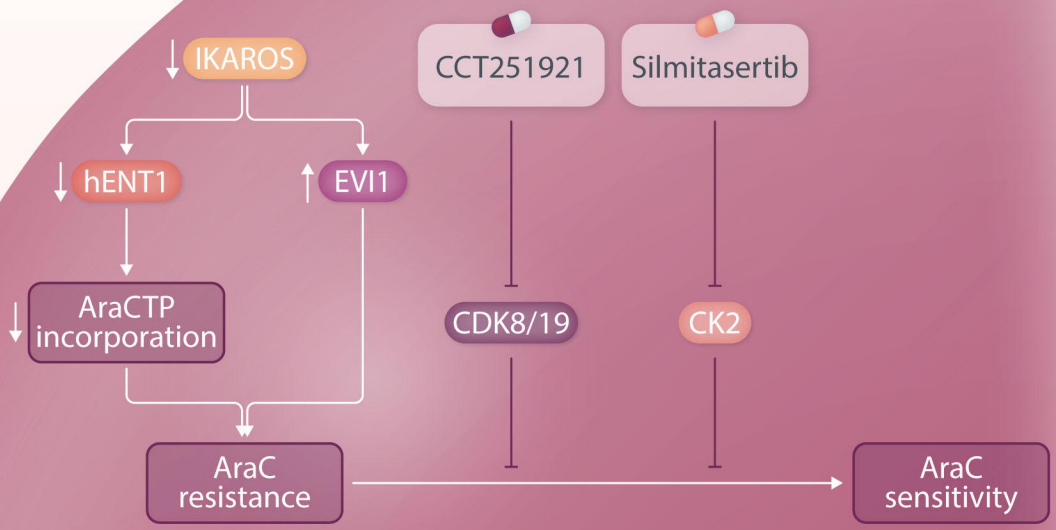
8 Mullighan CG, Su X, Zhang J, et al. Deletion of IKZF1 and prognosis in acute lymphoblastic leukemia. N Engl J Med. 2009;360(5):470-480.

9 Butler M, Vervoort BMT, van Ingen Schenau DS, et al. Reversal of IKZF1-induced glucocorticoid resistance by dual targeting of AKT and ERK signaling pathways. Front Oncol 2022;12:905665.

10 Song C, Ge Z, Ding Y, et al. IKAROS and CK2 regulate expression of BCL-XL and chemosensitivity in high-risk B-cell acute lymphoblastic leukemia. Blood. 2020;136(13):1520-1534.

**Figure Legend;**

**Figure 1.** Vervoort et al. demonstrate that IKZF1 deletion results in resistance to AraC via reduced expression of hENT1 and increased expression of Evi1. Treatment with CDK8/19 or CK2 inhibitors restores sensitivity to AraC treatment.


IKAROS
hENT1
EVI1
AraCTP incorporation
CCT251921
Silmitasertib
CDK8/19
CK2
AraC resistance
AraC sensitivity