

AMP case report: Adult B-lymphoblastic leukemia/lymphoma, *BCR-ABL1*-like

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Audra Kerwin, MD

Devon Chabot-Richards, MD

Laura Toth, DO, MPH

April 2021—A 71-year-old female with a history of asthma and hypertension initially presented to her local hospital complaining of shortness of breath. She was found to be pancytopenic with severe anemia (hemoglobin 5 g/dL). She was subsequently transferred to a tertiary care facility for further evaluation.

Bone marrow biopsy revealed a hypercellular marrow composed of 72 percent blasts (**Fig. 1**). Flow cytometric analysis revealed a B-lymphoblast immunophenotype with expression of CD34, dim CD45, CD19, CD79a, CD22, HLA-DR, TdT, CD200, CD33, and dim CD13. The blasts were negative for MPO, CD3, CD4, CD7, CD8, CD10, CD20, CD14, CD56, CD64, and CD117. Cytogenetic analysis identified a complex female karyotype exhibiting extensive numerical and structural abnormalities, including interstitial deletion of the long arm of chromosome five and an additional copy of *BCR* (22q11.2) (**Fig. 2**).

As these findings did not fulfill the diagnostic criteria for a subcategory of B-lymphoblastic leukemia/lymphoma (B-ALL) with recurrent genetic abnormality, screening for a *BCR-ABL1*-like RNA expression profile was performed using a low-density array card (commercially available from TriCore Reference Laboratories). This was positive for a *BCR-ABL1*-like profile with low *CRLF2* expression (**Fig. 3**), consistent with *BCR-ABL1*-like B-ALL not due to *CRLF2* rearrangement. Reflex FISH was negative for rearrangements involving *CRLF2*, *ABL1*, *ABL2*, *CSF1R*, *EPOR*, and *PDGFRB*. Further DNA and RNA sequencing was considered to identify the specific genetic alteration causing the *BCR-ABL1*-like profile. However, the oncologist decided to proceed with treatment for acute lymphoblastic leukemia, *BCR-ABL1*-like.

Given the diagnosis of *BCR-ABL1*-like B-ALL, the patient was treated with an aggressive chemotherapeutic regimen that included the addition of inotuzumab and intrathecal methotrexate. After induction, repeat bone marrow showed the presence of residual disease with a blast count of 8.3 percent. The patient eventually achieved morphologic remission after three months of intensive chemotherapy. Seven months later she returned with symptoms including fatigue and shortness of breath. Bone marrow biopsy at that time demonstrated dysplastic features and increased blasts of myeloid lineage (CD34, CD33, CD13, CD117, and subset MPO positive by flow cytometric analysis, negative for CD19, CD20, and TdT), and a diagnosis of therapy-related myeloid neoplasm (t-MN) was rendered (**Fig. 4**).

Cytogenetic analysis revealed a karyotype with shared abnormalities to that of the patient's original *BCR-ABL1*-like B-ALL, including the interstitial deletion of the long arm of chromosome five (**Fig. 5**). This may indicate a clonal relationship between the two, including underlying clonal hematopoiesis or the rare phenomenon of lineage switching. In this event, leukemic cells immunophenotypically and morphologically convert to a different lineage. The exact biological mechanism underlying lineage switch remains elusive, but molecular and cytogenetic analysis of these events has uncovered interesting findings that may indicate therapy-related clonal selection.¹ However, this rare yet well-described event typically occurs in a short time frame, immediately after or even during induction therapy. Thus, the diagnosis of t-MN was rendered.¹ The patient died three months later.

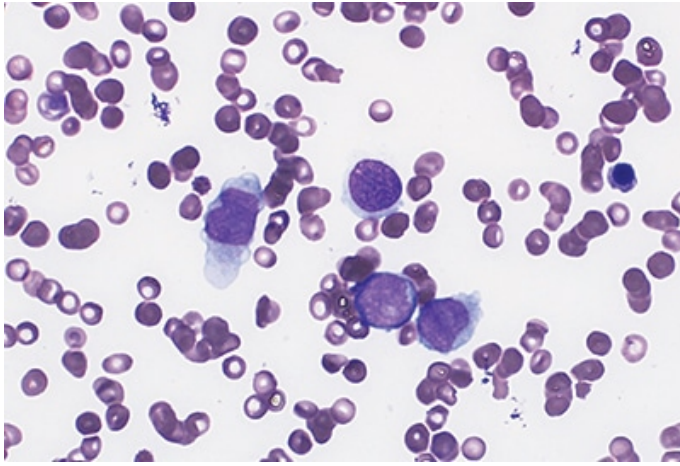


Fig. 1. Bone marrow aspirate at diagnosis of ALL shows blasts with large size, high nuclear-to-cytoplasmic ratio, and prominent nucleoli (Wright stain, 40×).

B-lymphoblastic leukemia/lymphoma, *BCR-ABL1*-like. The category of B-lymphoblastic leukemia/lymphoma (B-ALL) with recurrent genetic abnormalities has expanded over time and is now composed of nine distinct entities in the latest revision of the World Health Organization *Classification of Tumours of Haematopoietic and Lymphoid Tissues*. The update includes two new diagnostic subcategories: B-ALL with intrachromosomal amplification of chromosome 21 and B-ALL, *BCR-ABL1*-like.² *BCR-ABL1*-like ALL is a genetically heterogeneous group of diseases showing gene expression profiles similar to B-ALL with the *BCR-ABL1* translocation. These leukemias often carry translocations of other tyrosine kinase genes as well as *CRLF2* and *EPOR*. Seen in 10 to 20 percent of pediatric ALL, the incidence of *BCR-ABL1*-like increases with age, reaching 20 to 30 percent in adult ALL. Although originally described in pediatric patients, identification is of particular importance in adults because it portends a significantly worse overall and event-free survival, with a five-year survival of approximately 23 percent.^{3,4}



Fig. 2. Karyogram at the initial time of diagnosis with a composite karyotype of 46,XX,del(5)(q22q35)[2]/44-46,idem,del(8)(q11.2),add(11)(p15),del(16)(p12),-18,-20,-21,+22,add(22)(p11.1),add(22)(p11.1),+mar[cp17]/46,XX[1]

The diagnosis of *BCR-ABL1*-like ALL is unique in that it lacks a defining single

and/or discrete cytogenetic or molecular derangement. Although it lacks the t(9;22)(q34.1;q11.2), *BCR-ABL1* gene fusion, it has a “gene expression pattern very ‘similar’ to that seen in B-ALL with *BCR-ABL1*.”^{5,6} Over 60 different rearrangements and mutations giving rise to a *BCR-ABL1*-like profile have been identified.⁶ These aberrations most commonly cause activation of the JAK/STAT pathway or the ABL class of genes, leading to increased kinase signaling.⁶

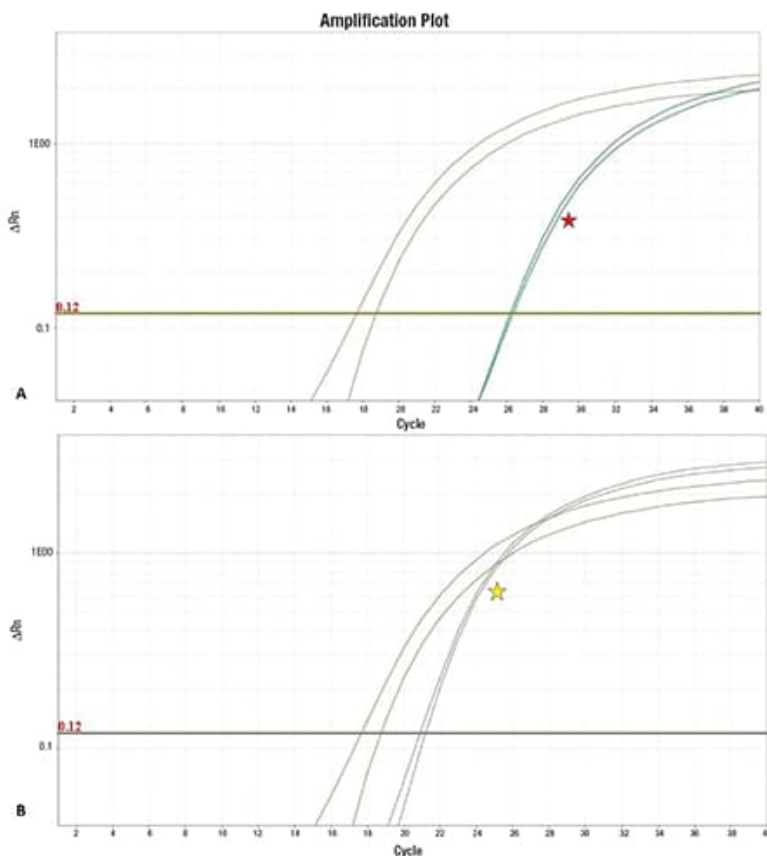


Fig. 3. The determination of a Ph-like profile is made via a low-density array, which calculates a composite RNA expression score for a set of 15 genes based on the PCR cycle when signal is detected compared with a control gene (olive green). A strict numerical cutoff for determining increased expression of each individual gene cannot be provided because each gene is internally normalized for each specific case (further description can be found in the supplemental information of reference 10). **A)** One gene evaluated is *CRLF2*, which was low as shown above (red star). **B)** A second gene evaluated, *IGJ*, shows relatively increased expression (yellow star).

The most commonly reported abnormality is rearrangement of the *CRLF2* gene leading to increased expression (30 to 50 percent of cases).⁶ Common translocation partners include IgH and *P2RY8*.⁷ Additionally, 30 to 50 percent of these *CRLF2*-rearranged cases harbor a concurrent *JAK1* or *JAK2* activating mutation that may be amenable to targeted therapy with agents such as ruxolitinib in combination with conventional chemotherapy.⁶ Additional targetable rearrangements associated with a *BCR-ABL1*-like profile include *EPOR* and the ABL class of genes: *ABL1*, *ABL2*, *CSF1R*, *PDGFRA*, and *PDGFRB*, which may be responsive to the addition of dasatinib or imatinib.^{3,4,7,8} Many of

these rearrangements can be evaluated using FISH. Additional fusions and mutations leading to increased kinase signaling, including *IKZF1*, *FGFR1*, and *RAS*, have been less commonly reported.⁴

Based on current guidelines, patients who should undergo *BCR-ABL1*-like screening should include all children diagnosed with B-ALL who fall into the high-risk category, other than those with the t(9;22)(q34.1;q11.2) *BCR-ABL1* and t(12;21)(p13.2;q22.1) *ETV6-RUNX1*, which exclude the diagnosis *BCR-ABL1*-like B-ALL. Children with standard-risk B-ALL may be considered for screening if there is residual disease after induction, or if there is central nervous system or testicular involvement.⁶ Finally, as in this case, adults with B-ALL who are negative for the t(9;22)(q34.1;q11.2), *BCR-ABL1* gene fusion should also be screened.⁶

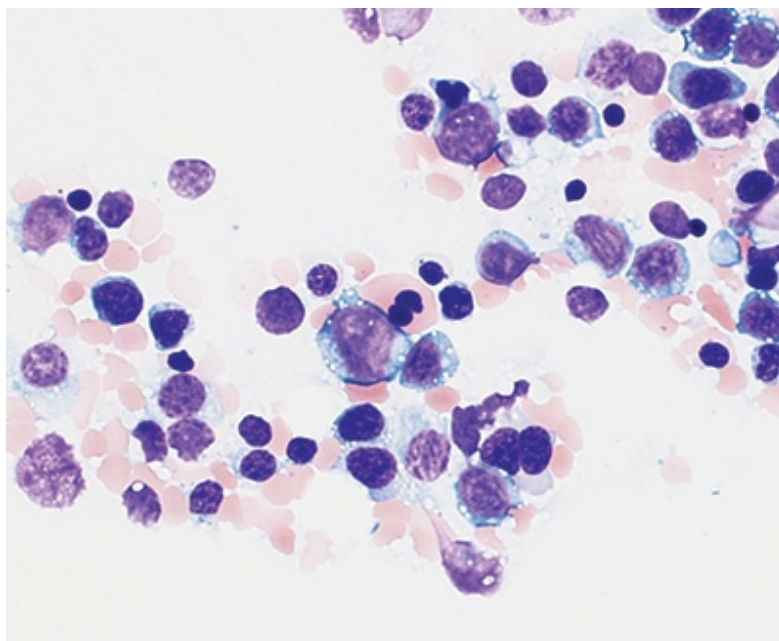


Fig. 4. Bone marrow aspirate at diagnosis of t-MN shows blasts with intermediate to large size, cytoplasmic vacuoles, moderate nuclear-to-cytoplasmic ratio, and prominent nucleoli. There is dyserythropoiesis with multinucleation and megaloblastic changes (Wright stain, 40×).

In the absence of gene expression profiling, diagnosis of *BCR-ABL1*-like ALL may be time-consuming and costly due to the wide number of translocations and mutations seen in this entity. However, identification is of the utmost importance for prognostic and therapeutic purposes. Common strategies include testing with a limited FISH panel, often starting with probes to identify *CRLF2* rearrangements and reflexing negative cases to analysis with additional FISH probes.⁹ Flow cytometry assays to assess for high *CRLF2* expression are also available. These approaches are specific but not sensitive, as they do not identify the wide range of genetic alterations associated with this disease. Additionally, large next-generation sequencing panels to evaluate for a broad range of possible mutations and RNA fusions are available, though these large panels are costly and can have a turnaround time upward of three weeks.



Fig. 5. Karyogram of the patient's therapy-related myeloid neoplasm. The final karyotype was 46,XX,del(5)(q22q35)[1]/47,idem,del(3)(q12),-7,der(7)ins(7;?)(q11.2;?),+8,del(16)(q12),+mar[7]/46,XX[4]. The del(5) abnormality is shared between the cytogenetic findings at diagnosis of B-ALL and at diagnosis of t-MN.

Quantitative reverse transcriptase-PCR-based low-density array (LDA) provides a sensitive and rapid approach to identify cases of *BCR-ABL1*-like ALL including those with abnormalities outside of the commonly used FISH panels. The LDA assay was developed by the Children's Oncology Group and evaluates the expression levels of 15 previously described genes (and a control gene) shown to be associated with the diagnosis *BCR-ABL1*-like ALL.¹⁰⁻¹³ A score is generated based on a composite of the individual amplification curves to determine if the case shows expression levels at or above the validated threshold of positivity.¹⁰ A coefficient of 0.5-1 is considered positive for expression of a Ph-like profile.¹⁰ The assay also screens for multiple fusions such as *BCR-ABL1* and *ETV6-RUNX1* that would preclude the diagnosis of *BCR-ABL1*-like ALL and those that are commonly associated with *BCR-ABL1*-like ALL such as *P2RY8-CRLF2*.¹⁰⁻¹³ Cases with positive LDA screens can be reflexed to further testing, as specific mutations and less common fusion rearrangements cannot be identified using this technology. Importantly, this screening method allows clinicians to risk stratify appropriately, develop a treatment plan, and possibly enroll patients into clinical trials as additional tests (FISH and/or sequencing) are completed.

BCR-ABL1-like B-ALL is a recently described entity, and thus significant studies describing treatment effects in adult patients are lacking. Patients often receive more intensive treatment, which may be linked to development of t-MN.¹⁴ Development of a secondary hematologic malignancy following treatment underscores the prognostic implications of *BCR-ABL1*-like B-ALL, including the hope that therapy targeting the aberrant genes or pathways may effect better outcomes.

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Dr. Kerwin is a hematopathology fellow, Dr. Chabot-Richards is associate professor, and Dr. Toth is visiting assistant professor, Department of Pathology, University of New Mexico, Albuquerque.

Test yourself answers

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1. Which of these patients with B-lymphoblastic leukemia (B-ALL) should be screened for a Philadelphia-like (Ph) expression profile?

- a. Pediatric patient with new diagnosis and karyotype showing hyperdiploidy.
- b. Pediatric patient with new diagnosis and karyotype showing *KMT2A* rearrangement.
- c. Pediatric patient with morphologic remission at end of induction chemotherapy.
- d. **Adult patient with new diagnosis and karyotype negative for t(9;22) *BCR-ABL1*.**

2. What is the prognostic and therapeutic significance of Ph-like B-ALL?

- a. Favorable prognosis; may respond to inotuzumab.
- b. Favorable prognosis; may respond to immunotherapy.
- c. **Worse prognosis; may respond to tyrosine kinase inhibitors.**
- d. Worse prognosis; may respond to immunotherapy.

3. What is the most common molecular abnormality associated with Ph-like B-ALL?

- a. ***CRLF2* gene rearrangements.**
- b. *EPOR* gene rearrangements.
- c. Activating *JAK1* or *JAK2* point mutations.
- d. *KMT2A* rearrangement; t(4;11) or *KMT2A/AF4* fusion.