EXCEPTIONAL CASE REPORT

blood advances

Larotrectinib in TRK fusion–positive pediatric B-cell acute lymphoblastic leukemia

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Key Points

- Larotrectinib can be effective in patients with ETV6-NTRK3–positive B-cell lymphoblastic leukemia, inducing prolonged molecular remission.
- Single-agent tyrosine kinase inhibitor treatment could be a valuable treatment option in subgroups of kinase fusion–positive ALL patients.

Introduction

Rearrangements involving neurotrophic receptor tyrosine kinase (*NTRK*) genes can generate fusion oncoproteins driving tumor development and survival.[1] *NTRK* gene fusions have been identified across a range of adult and pediatric solid malignancies.[2] B-cell acute lymphoblastic leukemia (ALL) can harbor an *ETV6-NTRK3* gene fusion in ~1% of the so-called "Philadelphia-like" cases.[3] *ETV6-NTRK3* fusion–positive B-cell ALL is characterized by rapid proliferation and infiltration of the central nervous system (CNS) in preclinical models.[4] Previously published phase 1 data for larotrectinib (NCT02637687), a highly selective tropomyosin receptor kinase (TRK) inhibitor, has shown a 93% response rate and good tolerability in a cohort of pediatric patients with TRK-positive relapsed/refractory solid tumors, excluding leukemia patients.[5] Adverse events included increased alanine and aspartate aminotransferase elevations, leucopenia, decreased neutrophil count, and vomiting, with no grade 4 or grade 5 events. Pharmacokinetics revealed that a dose of 100 mg/m^2 (with a maximum dose of 100 mg) twice a day resulted in a similar exposure as in adults treated with the recommended phase 2 dose of 100 mg twice a day. Exposure in the CNS was also confirmed.[5]

Here, we report the successful use of larotrectinib in a child with a relapse of *ETV6-NTRK3* fusion positive B-cell ALL early after hematopoietic allogeneic stem cell transplantation (HSCT).

Case description

A 6-year-old boy with B-cell ALL and CNS infiltration (National Cancer Institute high risk) had received polychemotherapy according to the high-risk arm of the Associazione Italiana Ematologia Oncologia Pediatrica-Berlin-Frankfurt-Muenster (AIEOP-BFM) 2009 protocol (NCT01117441). At diagnosis, conventional cytogenetics and fluorescence in situ hybridization analyses were performed in a peripheral blood sample with hyperleukocytosis (590 000 leukocytes per microliter).[6] For the detection of breakpoints affecting the *ETV6* locus, a commercially available *ETV6* break-apart probe was applied. The aberrant karyotype at diagnosis pointed to an unbalanced translocation of the *ETV6* locus (47,XY,+15[19].nuc ish 12p13(5′ETV6x4)(3′ETV6-x2)(5′ETV6 con 3′ETV6x2)[85/100]). The patient exhibited poor response to prednisone and persistent high levels of minimal residual disease (MRD; 10^{-3}) after consolidation. Therefore, allogeneic HSCT was performed according to protocol recommendations. Before HSCT, MRD in the bone marrow was negative. The conditioning regimen consisted of total body irradiation (12 Gy) and etoposide (60 mg/kg). The transplant was obtained from a matched unrelated donor. For graft-versus-host disease prophylaxis, anti-thymocyte globulin, cyclosporine A, and methotrexate were given.[7] The patient suffered from limited acute skin graft-versus-host disease (stage I; overall clinical grade 1[8]) upon engraftment. Fifty days post-HSCT, the patient relapsed (ocular, CNS, bone marrow), and immunosuppressive therapy was stopped (day 0 in Figure 1A; supplemental Figure 1). At relapse, RNA-sequencing was performed on the initial sample, and the cytogenetically cryptic *ETV6-NTRK3* fusion was detected. The relapse could be initially salvaged

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The full-text version of this article contains a data supplement.

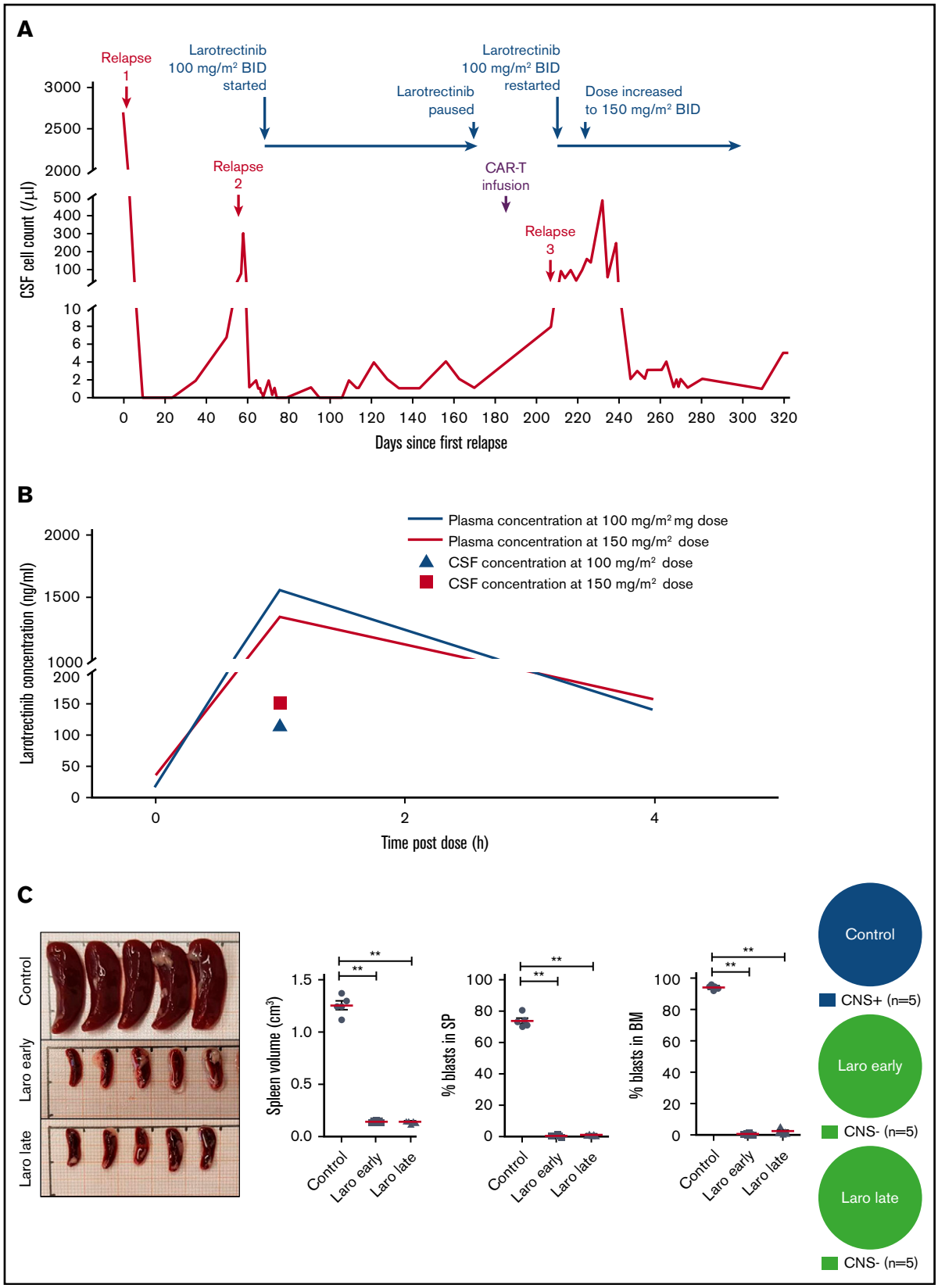

Figure 1. Patient and patient-derived xenograft data on Larotrectinib in a pediatric B-cell ALL patient. (A) Treatment course and cerebrospinal fluid (CSF) cell count over time. The day of first relapse post-HSCT (day 50 post-HSCT) was set as day 0. (B) Larotrectinib plasma and CSF concentrations. (C) Preclinical efficacy of larotrectinib initiated at 2 time points (day +1 postinjection ["laro early"] and day +8 postinjection ["laro late"]) in an NSG xenograft model (n = 5 mice per group). Images of spleens, spleen volumes, infiltration of bone marrow (BM) and spleens (SP) by flow cytometry, and CNS infiltration by morphology. $^{**}P < .0001$.

by blinatumomab combined with intrathecal chemotherapy. At day 55, the patient relapsed again (overt CNS, MRD 10^{-3} in the bone marrow), and larotrectinib was initiated at 100 mg/m^2 twice a day as part of an expanded access program for patients ineligible for an ongoing larotrectinib clinical trial (NCT03025360), in combination with intraventricular chemotherapy with etoposide (1 mg Monday and Thursday) and methotrexate (2 mg Tuesday, Wednesday, and Thursday) alternating weekly (Figure 1A; supplemental Figure 1). This resulted in molecular remission in the CNS and the bone marrow at day 121, as measured by polymerase chain reaction for immunoglobulin/T-cell receptor genes (Figure 1A; supplemental Figure 1). Larotrectinib treatment was subsequently stopped for T-cell apheresis (day 170; Figure 1A), and chimeric antigen receptor (CAR) T-cell therapy (tisagenlecleucel) was administered on day 190. The patient went into B-cell aplasia, indirectly suggesting efficacy of the CAR T cells. However, the patient again relapsed in the CNS shortly after CAR T-cell infusion (bone marrow remained MRD negative), which may be due to impaired penetration of CAR T cells into the CNS. Larotrectinib was restarted on day 215. In addition, the same CNS-directed chemotherapy was reinitiated. The larotrectinib dose was increased to 150 mg/m^2 twice a day to improve CNS compartment penetration confirmed using pharmacokinetic measurements (Figure 1B). The patient again achieved MRD negativity in the CNS and bone marrow on day 281. Intervals for intraventricular etoposide and methotrexate are currently 1 week on and 1 week off. The patient remains in molecular remission (day 320). There have been no obvious adverse events related to larotrectinib. In addition, patient-derived xenograft data in mice were obtained with material from this patient. Mice were injected with leukemia cells, and larotrectinib treatment was initiated on day +1 ("laro early") or on day +8, after detection of >1% human leukemic cells in the blood of the animals ("laro late"). All mice were euthanized on day +25 to assess leukemic burden in different organs, which was significantly lower in all compartments in larotrectinib-treated mice compared with controls (Figure 1C; supplemental Figure 2). This was independent of the time at which larotrectinib was initiated (Figure 1C; supplemental Figure 2) and suggests that larotrectinib was also efficient in a preclinical model with cells from this patient.

Methods

The detection of the *ETV6-NTRK3* fusion, MRD analyses in bone marrow and cerebrospinal fluid samples, and the preclinical in vivo experiment with larotrectinib are described in supplemental Methods. Ethics committee approval has been obtained for a single-patient protocol.

Results and discussion

Larotrectinib has the potential to be an effective treatment for patients with *ETV6-NTRK3*–positive B-cell ALL, including patients with CNS infiltration. This patient demonstrates a clinical and a quantifiable response consistent with previous reports of larotrectinib therapy. We were able to treat 2 relapses in the same patient with larotrectinib and brought the patient into continued molecular remission in all compartments at the present time. This is striking because the patient had received all lines of conventional therapy (including allogeneic HSCT, antibody therapy with blinatumomab, and CAR T cells), and all were inefficient. Chemotherapy was performed in the CNS compartment only; therefore, we conclude that MRD negativity in the bone marrow is primarily due to larotrectinib, and CAR T-cell therapy may also have contributed. In the CNS, a part of the antileukemic effect is certainly mediated by the low-dose chemotherapeutic agents. We are planning to increase the chemotherapy pauses to ultimately withdraw intraventricular chemotherapy. We show for the first time that treatment with a specific tyrosine kinase inhibitor, with very little additional therapy, can sustain molecular remission in a patient with a very aggressive subtype of acute leukemia, which is an exceptional finding. Additionally, the remarkable clinical response in our patient supports the unusual preclinical efficacy of larotrectinib in published models[4] and in our own. Furthermore, our treatment results in this patient provide an argument for incorporating sequencing approaches detecting potentially targetable lesions and gene fusions into the diagnostics of ALL, at least in patients with high-risk disease features, resistance to therapy, or relapse. Preclinical studies have assessed other TRK inhibitors, including crizotinib (traditionally used as an inhibitor of anaplastic lymphoma receptor tyrosine kinase) and entrectinib in *ETV6-NTRK3* fusion B-cell ALL models.[3,9] Also, next-generation TRK inhibitors are currently in phase 1 studies for solid tumors (eg, LOXO-195 [NCT03215511] and TPX-0005 [NCT03093116]) in patients who have failed first-generation inhibitors, opening further avenues in the future. Because TRK fusion–positive leukemia is a rare entity, precluding the performance of larger clinical trials, it is important to show treatment responses in single cases of leukemia patients.

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Authorship

Contribution: D.M.S., A.V.R. and G.C. treated the patient; D.M.S. and G.C. initiated larotrectinib treatment; L.L., F.V., and D.W. contributed xenograft data; S.B. and A.K.B. provided diagnostic data; M.C.C. and D.H. provided guidance in treating the patient with larotrectinib and contributed pharmacokinetics data; and all authors wrote, discussed, and approved the manuscript.

Conflict-of-interest disclosure: M.C.C. was an employee of Loxo Oncology, Inc., a wholly owned subsidiary of Eli Lilly and Company during the time this work was completed; is a stockholder of Bayer AG; and has a patent licensed to Loxo Oncology, Inc. (62/318041). D.H. is an employee of Loxo Oncology, Inc., a wholly owned subsidiary of Eli Lilly and Company. The remaining authors declare no competing financial interests.

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