**ETV6-FLT3**–positive myeloid/lymphoid neoplasm with eosinophilia presenting in an infant: an entity distinct from JMML

Barbara Spitzer,1,2 Filemon S. Dela Cruz,1 Glorymar D. Ibanez Sanchez,3 Yanming Zhang,4 Wenbin Xiao,4 Ryma Benayed,4 Alina Markova,5,6 M. Irene Rodriguez-Sanchez,1 Nancy Bouvier,1 Mikhail Roshal,4 Andrew L. Kung,1 and Neerav Shukla1

1MSK Kids, Memorial Sloan Kettering Cancer Center, New York, NY; 2Department of Pediatrics, Weill Cornell Medicine, New York, NY; 3Sloan Kettering Institute, 4Hematopathology Service, Department of Pathology, and 5Dermatology Service, Department of Medicine, Memorial Sloan Kettering Cancer Center, New York, NY; and 6Department of Dermatology, Weill Cornell Medicine, New York, NY

---

**Key Points**

- Rare cases of fusion-driven myeloproliferative neoplasms may mimic more common presentations, such as JMML in this case.
- Identifying these fusions will often lead to a change in therapy, using tyrosine kinase inhibitors to induce faster and deeper remissions.

---

**Myeloid/lymphoid neoplasm with eosinophilia (MLN-Eo) is a World Health Organization (WHO) established category of hematologic malignancies primarily arising in adults. We discuss an 8-month-old infant who presented with clinical features similar to those of juvenile myelomonocytic leukemia (JMML) but who was diagnosed with MLN-Eo driven by an ETV6-FLT3 fusion. Results of patient-derived leukemia ex vivo studies demonstrated increased sensitivity to type I FLT3 inhibitors as compared with type II inhibitors. Treatment with the type I inhibitor gilteritinib resulted in complete immunophenotypic and cytogenetic remission. This patient subsequently underwent a hematopoietic stem cell transplant and remains in complete remission 1 year later. This is the youngest patient reported with an ETV6-FLT3 fusion and adds to the mounting reports of FLT3-rearranged MLN-Eo, supporting its addition to the WHO classification. Furthermore, this case highlights the clinical utility of ex vivo drug testing of targeted therapies.**

**Introduction**

We present the first report of an infant with an ETV6-FLT3 fusion-driven myeloid/lymphoid neoplasm with eosinophilia, T-lymphoid, and myeloid differentiation (MLN-Eo), which exhibited unique sensitivity to type I FMS-like tyrosine kinase 3 (FLT3) inhibitors.

**Case description and results**

The study was conducted in accordance with the Declaration of Helsinki. An 8-month-old male infant presented to a local emergency department with a 2-week history of fever, irritability, and decreased oral intake. On examination, the patient was found to be febrile, with massive hepatosplenomegaly, lymphadenopathy, and a diffuse rash (Figure 1A). A complete blood count (CBC) showed a white blood count (WBC) of $250 \times 10^3/\mu L$, with eosinophilia (23%), circulating promonocytes (7%), and blasts (14%). Bone marrow aspirate was performed, and a preliminary diagnosis of juvenile myelomonocytic leukemia (JMML) was made. After leukapheresis and initiation of hydroxyurea, the patient was transferred to Memorial Sloan Kettering Cancer Center for further workup and management.

On transfer to our center, the aforementioned physical findings were observed; the patient was also noted to have significant edema and associated respiratory distress. A CBC on arrival revealed a WBC of $68 \times 10^3/\mu L$. Flow cytometric analysis of peripheral blood revealed the following: an abnormal immature T-cell population (12%), an abnormal myeloid blast population (0.1%), monocytes (17%), and eosinophilia (21%). Review of submitted bone marrow aspirate smears confirmed increased blasts/blast equivalent (23% in total) but no significant dysplasia (Figure 1B). Cytogenetics (Figure 1C) revealed a clonal t(12;13)(p13;q12) by karyotype, confirmed by fluorescence in situ hybridization (FISH).
as a rearrangement involving *ETV6* in 83% of cells. The fusion was identified in multiple flow-sorted cell lineages, including CD34+ myeloid blast, immature T-cell, and monocyte populations, at high levels. Targeted DNA- and RNA-sequencing revealed an *ETV6*-*FLT3* fusion, fusing *ETV6* exons 1-6 to *FLT3* exons 14-24 (Figure 1D) and no evidence of RAS pathway mutations. Based on these findings, a diagnosis of MLN-Eo was made. Results of the rash skin biopsy confirmed involvement by the same leukemic process, containing the same *ETV6-FLT3* fusion as in the blood and marrow.

Given the critical status of the patient, sorafenib (150 mg/m² per dose, twice daily), targeting the *FLT3* fusion, was started to provide clinical stabilization. After 12 days of sorafenib and hydroxyurea therapy, there was marked improvement of hepatosplenomegaly and leukemia cutis, resolution of respiratory distress, and improvement of leukocytosis to a WBC of 12 × 10⁹/μL. However, evaluation of the bone marrow revealed residual leukemia with 13% blasts morphologically and 57% *ETV6*-rearranged cells by FISH. The patient went on to receive 3 cycles of intensive chemotherapy combined with continuous sorafenib (Figure 2A). This regimen led to reduction in disease burden as shown by a decrease in FISH positivity for the *ETV6* rearrangement after each cycle of chemotherapy. However, after completion of these 3 cycles of chemotherapy and continuous sorafenib, bone marrow evaluation revealed persistent low-level minimal residual disease (MRD) by FISH and targeted RNA-sequencing.

During treatment with combination chemotherapy and sorafenib, patient-derived leukemia cells from diagnosis were treated with a panel of *FLT3* inhibitors for 96 hours and 50% inhibitory concentration values calculated following colorimetric (alamarBlue, MilliporeSigma) assessment of viability (Figure 2B). Relatively lower 50% inhibitory concentration values were observed across type I *FLT3* inhibitors compared with type II inhibitors, suggesting preferential sensitivity to type I *FLT3* inhibition. Based on these results, gilteritinib (a type I inhibitor) was started with a goal to achieve MRD-negative remission. After 2 months of single-agent gilteritinib, the patient had no detectable rearrangement of *ETV6* by FISH. He then underwent a conventional bone marrow transplant (BMT) from his HLA-identical brother. He developed stage III skin graft-versus-host disease, which responded rapidly to treatment. The patient restarted gilteritinib on day +45 post-transplant. All marrow evaluations after BMT have been negative for *ETV6*.
rearrangement by FISH. Furthermore, his most recent marrow evaluation was negative for ETV6-FLT3 by ddPCR of bone marrow aspirate–derived DNA (Figure 2B). The patient completed 1 year of posttransplant therapy with gilteritinib and remains free of disease.

Discussion

MLN-Eo and rearrangement of PDGFRα, PDGFRβ, or FGFR1, or with PML-1-JAK2, is a World Health Organization–established category of hematologic malignancies primarily arising in adults. The oncogenic kinase fusion in this case was defined by the joining of exon 6 of ETV6 to exon 14 of FLT3. The FLT3 product retains the juxtamembrane and both tyrosine kinase domains. In addition, FLT3–fusion MPN has shown in vitro and in vivo sensitivity to FLT3 inhibitors. We started our patient on the type II FLT3 inhibitor sorafenib based on safety and efficacy data from a Children’s Oncology Group study evaluating its efficacy in children with FLT3–internal tandem duplication–positive acute myeloid leukemia. Although we were able to reduce disease burden down to an MRD-positive level with sorafenib and chemotherapy, it was only after treatment with the type I inhibitor gilteritinib that we were able to achieve MRD negativity according to FISH.

In conclusion, this infant presented with clinical features suggesting JMML but was ultimately identified to have a tyrosine kinase fusion–driven MLN-Eo. Our clinical experience, as well as others reported in the literature, suggests that a strategy incorporating tyrosine kinase inhibitors with hematopoietic stem cell transplantation is an effective treatment paradigm for this rare group of kinase-driven MPN. Ex vivo patient-specific data suggested preferential sensitivity to type I FLT3 inhibitors, which aided our selection of a more sensitive inhibitor and ultimately a cytogenetic complete response for our patient. Further studies are needed to better compare the genetic and biologic features of these entities compared with classic RAS pathway–driven JMML.

Acknowledgment

Funding support was provided in part by a National Institutes of Health, National Cancer Institute Cancer Center Support Grant (P30 CA008748).

Figure 2. Patient treatment and disease course and ex vivo studies of patient-derived leukemia. (A) Clinical treatment course (top axis) and disease burden, as represented by log(proportion of FISH+ cells) on the left y-axis and log(proportion of digital droplet polymerase chain reaction [ddPCR] + cells) on the right y-axis, throughout therapy (time on x-axis). (B) Results from ex vivo drug screen showed the lower IC50(g20) of type I compared with type II FLT3 inhibitors in this patient’s leukemia samples. C1, C2, C3, cycle 1, cycle 2, cycle 3; Inh, inhibitor.
Authorship

Contribution: B.S. and N.S. provided direct patient care, performed analyses, and wrote the paper; F.S.D.C. and G.D.I.S. designed and performed the experiment; Y.Z., W.X., M.R., and R.B. performed analyses and diagnostic evaluation and edited the paper; A.M. provided direct patient care and diagnosis; M.I.R.-S. and N.B. obtained clinical data and helped write the paper; and A.L.K. helped design the experiments and edited the paper.

Conflict-of-interest disclosure: W.X. has received research support from Stemline Therapeutics. R.B. has received a grant and travel credit from ArcherDx; honoraria for advisory board participation from Loxo Oncology; and speaking fees from Illumina. A.M. is on the advisory board of AstraZeneca; and receives research funding from Incyte. M.R. has equity in Auron Therapeutics; and has been involved with provision of services from Celgene and Physicians' Education Resource. The remaining authors declare no competing financial interests.

ORCID profiles: B.S., 0000-0003-1603-1381; W.X., 0000-0001-8586-8500.

Correspondence: Neerav Shukla, MSK Kids, 1275 York Ave, New York, NY 10065; e-mail: shuklan@mskcc.org.

References

1. Arber DA, Orazi A, Hasserjian R, et al. The 2016 revision to the World Health Organization classification of myeloid neoplasms and acute leukemia. Blood. 2016;127(20):2391-2405.

2. Zhang H, Paliga A, Hobbs E, et al. Two myeloid leukemia cases with rare FLT3 fusions. Cold Spring Harb Mol Case Stud. 2018;4(6):a003079.

3. Walz C, Erben P, Ritter M, et al. Response of ETV6-FLT3-positive myeloid/lymphoid neoplasm with eosinophilia to inhibitors of FMS-like tyrosine kinase 3. Blood. 2011;118(8):2239-2242.

4. Yu HA, Xinh PT, Masuda M, et al. FLT3 is fused to ETV6 in a myeloproliferative disorder with hyper eosinophilia and a t(12;13)(p13;q12) translocation. Leukemia. 2006;20(8):1414-1421.

5. Hosseini N, Craddock K, Salehi-rad S, et al. FLT3 /ETV6 fusion in a mixed-phenotype acute leukemia arising in lymph nodes in a patient with myeloproliferative neoplasm with eosinophilia. J Hematop. 2014;7(2):71-77.

6. Falchi L, Mehrotra M, Newberry KJ, et al. ETV6-FLT3 fusion gene-positive, eosinophilia-associated myeloproliferative neoplasm successfully treated with sorafenib and allogeneic stem cell transplant. Leukemia. 2014;28(10):2090-2092.

7. Chonabayashi K, Hishizawa M, Matsui M, et al. Successful allogeneic stem cell transplantation with long-term remission of ETV6/FLT3-positive myeloid/lymphoid neoplasm with eosinophilia. Ann Hematol. 2014;93(3):535-537.

8. Keene P, Mendelow B, Pinto MR, et al. Abnormalities of chromosome 12p13 and malignant proliferation of eosinophils: a nonrandom association. Br J Haematol. 1987;67(1):25-31.

9. Shao H, Wang W, Song J, et al. Myeloid/lymphoid neoplasms with eosinophilia and FLT3 rearrangement. Leuk Res. 2020;99:106460.

10. Jawhar M, Naumann N, Knut M, et al. Cytogenetically cryptic ZMYM2-FLT3 and DIAPH1-PDGFRB gene fusions in myeloid neoplasms with eosinophilia. Leukemia. 2017;31(10):2271-2273.

11. Stiegitz E, Taylor-Weiner AN, Chang TY, et al. The genomic landscape of juvenile myelomonocytic leukemia [published correction appears in Nat Genet. 2015;47(11):1333]. Nat Genet. 2015;47(11):1326-1333.

12. Chao AK, Meyer JA, Lee AG, et al. Fusion driven JMML: a novel CCDC88C-FLT3 fusion responsive to sorafenib identified by RNA sequencing. Leukemia. 2020;34(2):682-686.

13. Murakami N, Okuno Y, Yoshida K, et al. Integrated molecular profiling of myelomonocytic leukemia. Blood. 2018;131(14):1576-1586.

14. Moreiro C, Acquila M, Rosanda C, et al. HCMOGT-1 is a novel fusion partner to PDGFRB in juvenile myelomonocytic leukemia with t(5;17)(q33;p11.2). Cancer Res. 2004;64(8):2649-2651.

15. Niemeyer CM. JMML genomics and decisions. Hematology Am Soc Hematol Educ Program. 2018;2018:307-312.

16. Grand FH, Iqbal S, Zhang L, Russell NH, Chase A, Cross NC. A constitutively active SPTBN1-FLT3 fusion in atypical chronic myeloid leukemia is sensitive to tyrosine kinase inhibitors and immunotherapy. Exp Hematol. 2007;35(11):1723-1727.

17. Troade E, Dobbelstein S, Bertrand P, et al. A novel t(3;13)(q13;q12) translocation fusing FLT3 with GOLGB1: toward myeloid/lymphoid neoplasms with eosinophilia and rearrangement of FLT3? Leukemia. 2017;31(2):514-517.

18. Dafer N, Schlenk RF, Russell NH, Levis MJ. Targeting FLT3 mutations in AML: review of current knowledge and evidence. Leukemia. 2019;33(2):299-312.

19. Pollard JA, Alonzo TA, Brown PA, et al. Sorafenib in combination with standard chemotherapy for children with high allelic ratio FLT3/ITD+ AML improves event-free survival and reduces relapse risk: a report from the Children’s Oncology Group Protocol AAML1031. Blood. 2019;134(suppl 1):292.

20. Yao J, Xu L, Aybar U, et al. Myeloid/lymphoid neoplasms with eosinophilia/basophilia and ETV6-ABL1 fusion: cell-of-origin and response to tyrosine kinase inhibition. Haematologica. 2021;106(2):614-618.

21. Carl T, Patel A, Derman B, et al. Diagnosis and treatment of mixed phenotype (T-myeloid/lymphoid) acute leukemia with novel ETV6-FGFR2 rearrangement. Blood Adv. 2020;4(19):4924-4928.