Letter to the Editor
Diagnostic Hematology

Ann Lab Med 2017;37:77-80
https://doi.org/10.3343/alm.2017.37.1.77
ISSN 2234-3806 • eISSN 2234-3814

ANNALS OF LABORATORY MEDICINE

A Case of Chronic Myeloid Leukemia With Rare Variant *ETV6/ABL1* Rearrangement

Soo In Choi, M.D.[1,*], Mi-Ae Jang, M.D.[1,*], Woo Joon Jeong, M.T.[1], Byung Ryul Jeon, M.D.[1], Yong-Wha Lee, M.D.[1], Hee Bong Shin, M.D.[1], Dae-Sik Hong, M.D.[2], and You Kyoung Lee, M.D.[1]

Department of Laboratory Medicine and Genetics[1], Department of Internal Medicine[2], Division of Hematology & Oncology, Soonchunhyang University Bucheon Hospital, Soonchunhyang University College of Medicine, Bucheon, Korea

Dear Editor,

The translocation (9;12)(q34;p13) *ETV6/ABL1* rearrangement is a rare but recurrent chromosomal translocation associated with a variety of hematological malignancies, including CML, atypical CML, AML, and ALL [1]. The structure of the ETV6/ABL1 oncoprotein is similar to that of BCR/ABL1, and they initiate similar downstream pathways [2]. There are two *ETV6/ABL1* fusion isoforms: the type A isoform, which fuses *ETV6* exon 4 with *ABL1* exon 2; and the type B isoform, which fuses *ETV6* exon 5 with *ABL1* exon 2 [3, 4]. To date, 30 cases of *ETV6/ABL1* fusion have been reported [5, 6], and only one of these cases resulted in CML with positive *BCR/ABL1* rearrangement [7]. Herein, we report a rare case of CML with *ETV6/ABL1* rearrangement.

A 54-yr-old male was admitted with persistent leukocytosis. Complete blood counts showed a white blood cell count of 21.7 $\times 10^9$/L with 1% blasts, Hb of 126 g/L, and platelet count of 294 $\times 10^9$/L. Physical examination was unremarkable. Bone marrow (BM) analysis showed typical characteristics of CML (Fig. 1A, B). Chromosomal analysis of the BM cells demonstrated a balanced t(9;12)(q34;p13) translocation, which was not the Philadelphia chromosome (Fig. 1C). FISH analysis with probes for *BCR/ABL1* (Abbott Vysis, Des Plaines, IL, USA detected no fusion signal. However, reverse transcriptase (RT)-PCR analysis of the *BCR/ABL1* fusion transcripts yielded positive results; the reaction product was 700 bp long, indicating positive rearrangement and hence, presence of the P230 chimeric protein at the molecular level (Fig. 1D).

To visualize the *ETV6/ABL1* fusion signal, we prepared a mixture of two commercially available, locus-specific identifiers: a *BCR/ABL1* dual color, dual fusion translocation probe, and an *ETV6/RUNX1* extra signal dual color translocation probe (Abbott Vysis) (Fig. 1E, F). Metaphase and interphase FISH with the mixed *BCR/ABL1* and *ETV6/RUNX1* probes showed one yellow fusion signal at 9q34, which was derived from a green signal from *ETV6* and a red signal from *ABL1* (Fig. 1G, H). RT-PCR analysis of the *ETV6/ABL1* fusion transcript was positive for the 1,141-bp product, indicating a type B fusion (Fig. 1D). After diagnosis, the patient was transferred to another hospital, and therefore, follow-up BM examination was not possible.

ETV6/ABL1 rearrangement has been reported to result in enhanced tyrosine kinase activity and neoplastic transformation [3, 8]. A total of 13 cases of *ETV6/ABL1*-positive or atypical CML have been reported to date (Table 1) [5, 7]. Among those cases, including the present case, two were *BCR/ABL1* fusion-positive and 11 were either unknown or negative for the *BCR/ABL1* fusion. Both *BCR/ABL1* fusion-positive cases presented with per-

Received: April 20, 2016
Revision received: June 9, 2016
Accepted: August 25, 2016

Corresponding author: You Kyoung Lee
Department of Laboratory Medicine and Genetics, Soonchunhyang University Bucheon Hospital, Soonchunhyang University College of Medicine, 170, Jomaru-ro, Wonmi-gu, Bucheon 14584, Korea
Tel: +82-32-621-5941, Fax: +82-32-621-5944, E-mail: cecilia@schmc.ac.kr

*These two authors contributed equally to this work.

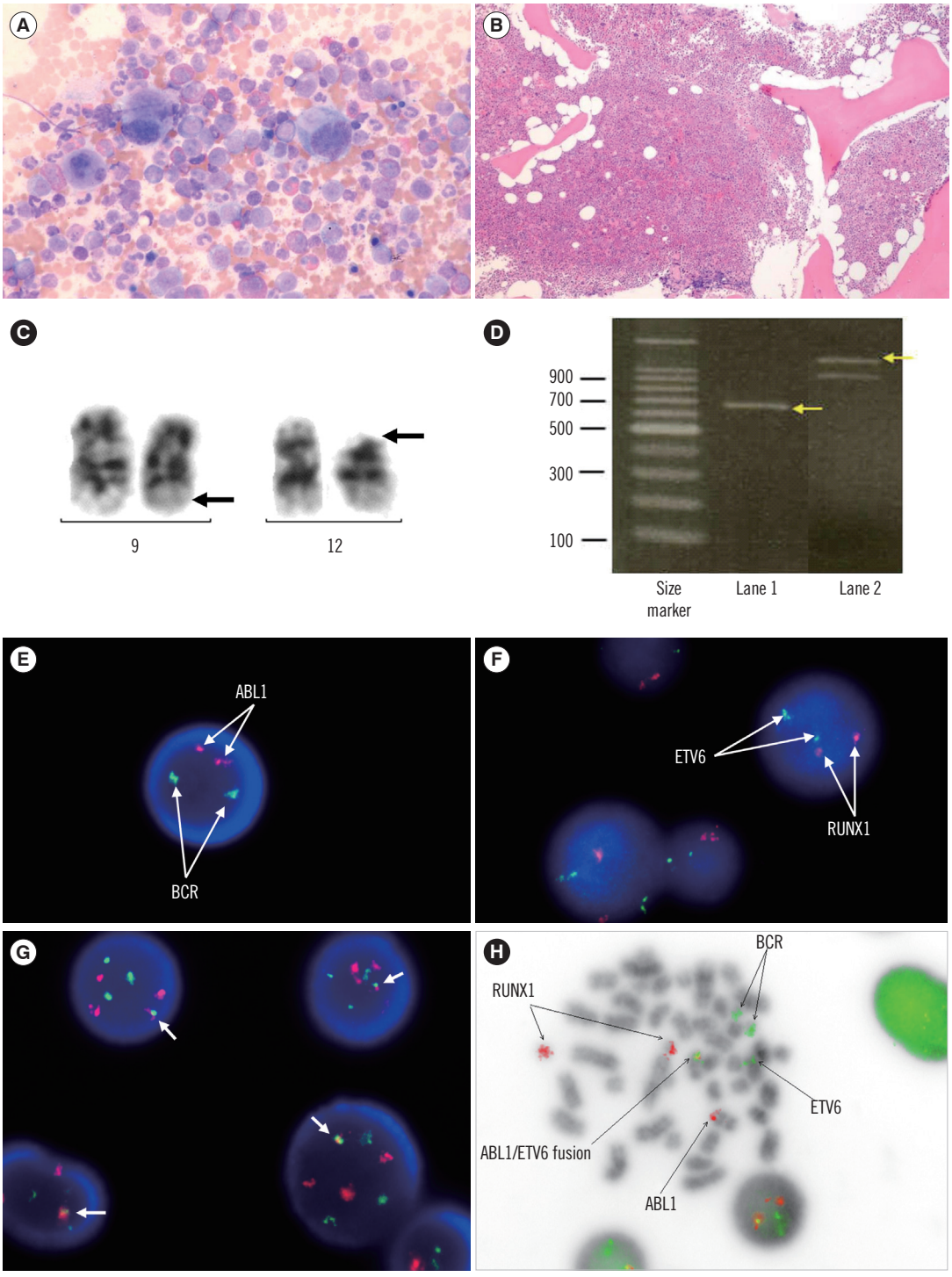

Fig. 1. Bone marrow (BM) aspiration, biopsy, karyotyping, reverse transcription (RT)-PCR, and FISH analyses of the present case. (A) BM aspiration (Wright-Giemsa stain, ×400) and (B) BM biopsy (hematoxylin & eosin stain, ×50) revealed 90% hypercellular marrow with a left-shifted neutrophilic series, an increased number of eosinophilic precursors, and small, hypolobated megakaryocytes. (C) Karyotyping showing t(9;12)(q34;p13); arrows indicate the translocated regions. (D) RT-PCR using *BCR/ABL1* and *ETV6/ABL1* primer pairs, revealing the 700-bp *BCR/ABL1* (lane 1, arrow) and 1,141-bp *ETV6/ABL1* (lane 2, arrow) fusion transcripts; left, 100-bp molecular weight marker ladder. The internal control 911-bp band is shown in lane 2. (E and F) FISH analysis using *BCR/ABL1* or *ETV6/RUNX1* probes, showing no abnormal signal. (G and H) FISH using the mixed *BCR/ABL1* and *ETV6/RUNX1* probes revealing one yellow fusion signal (*ABL1*, red; *ETV6*, green) on 9q34 analyzed in interphase (G) and metaphase (H) cells.

https://doi.org/10.3343/alm.2017.37.1.77

Table 1. Summary of patients with CML or atypical CML carrying the *ETV6/ABL1* fusion transcript

	Sex/Age (yr)	Splenomagaly	Initial CBC (WBC/Hb/Platelet)*	Eosinophilia	Karyotype	*BCR/ABL1* fusion (isoform)	*ETV6/ABL1* fusion isoform
1	NA/49	NA	NA/NA/NA	Yes	NA	NA	Type B
2	M/32	No	29/135/337	Yes	46,XY,t(12;14)(p12;q11-13)	Negative	Type B
3	M/59	Yes	27/66/344	Yes	46,XY,del(6)(p21),?t(9;12)(q34;p12)	NA	Type A
4	M/38	Yes	77.6/103/90	NA	46,XY	NA	Types A, B
5	M/53	No	22/131/378	Yes	46,XY	Negative	Types A, B
6	F/44	No	37/123/370	Yes	46,XX,t(9;12)(q34;p13)	Positive (P210)	NA
7	M/36	Yes	23.8/96/88	Yes	45,XY,-7,t(9;12)(q34;q13)	NA	Type B
8	M/72	NA	57/98/32	Yes	46,XY	NA	NA
9	F/24	Yes	98.8/113/261	Yes	46,XX	Negative	Type A
10	M/79	NA	35.2/141/176	Yes	46,XY	NA	NA
11	M/36	Yes	55/NA/NA	No	46,XY,t(9;12)(q34;p13)	Negative	NA
12	F/52	NA	Increased/NA/Increased	Yes	46,XX,t(9;12)(q34;p13)	NA	Types A, B
Present case	M/54	No	21.7/126/294	Yes	46,XY,t(9;12)(q34;p13)	Positive (P230)	Type B

Published cases were reviewed by Gancheva *et al* [5].
*Values are presented in the International System of Units (WBC, $\times 10^9$/L; Hb, g/L; Platelet, $\times 10^9$/L).
Abbreviations: CBC, complete blood count; WBC, white blood cell; F, female; M, male; NA, not available; RT-PCR, reverse transcription PCR.

sistent leukocytosis, eosinophilia, and no splenomegaly. Their pathological findings were consistent with those for CML, but *BCR/ABL1* rearrangement was not confirmed by karyotyping or FISH. Only RT-PCR revealed the rearrangement, and the amplicon size was 504 bp [7] and 700 bp in the present case, respectively. Marked eosinophilia, which is a common characteristic of the *ETV6/ABL1* translocation [7], was also predominant. Although the pathogenesis of eosinophilia is not clearly understood, *ETV6* is known to play an active role in the commitment of hematopoietic myeloid precursors to eosinophilic differentiation [9].

Rare cases of CML are associated with a *BCR* breakpoint that is considerably more directed towards the 3′ end than the major breakpoint cluster region, which encodes a P230 *BCR/ABL1* fusion protein. Our patient had a novel-sized *BCR/ABL1* fusion transcript (700 bp), which is ~140 bp smaller than the typically observed micro *BCR/ABL1* (c3a3) amplicon size of 838 bp, suggesting in-frame deletion of an exon. Although the lack of the Philadelphia chromosome observed by karyotyping and FISH is unusual, it is possible that RT-PCR is more sensitive than cytogenetics or FISH. Unfortunately, Sanger sequencing of the identified novel transcript could not be performed, and 2 weeks later, a repeat RT-PCR analysis of the peripheral blood failed to detect any *BCR/ABL1* fusion transcripts.

Detection of the *ETV6/ABL1* fusion may help to inform treatment plans for patients with rare hematologic malignancies. In these cases, tyrosine kinase inhibitors can be effective because of the significant overlap between the molecular targets of *ETV6/ABL1* and those of *BCR/ABL1* [10].

In conclusion, we identified an *ETV6/ABL1* translocation in a patient with CML, which was confirmed by FISH with combined *BCR/ABL1* and *ETV6/RUNX1* probes, as well as by RT-PCR analysis. This report will contribute to a better understanding of the clinical phenotype and molecular basis of this rare type of *ETV6/ABL1*-positive hematologic malignancy.

Authors' Disclosures of Potential Conflicts of Interest

No potential conflicts of interest relevant to this article were reported.

Acknowledgments

This work was supported by the Soonchunhyang University Research Fund.

REFERENCES

1. De Braekeleer E, Douet-Guilbert N, Morel F, Le Bris MJ, Basinko A, De Braekeleer M. ETV6 fusion genes in hematological malignancies: a review. Leuk Res 2012;36:945-61.
2. Malinge S, Monni R, Bernard O, Penard-Lacronique V. Activation of the NF-kappaB pathway by the leukemogenic TEL-Jak2 and TEL-Abl fu-

sion proteins leads to the accumulation of antiapoptotic IAP proteins and involves IKKalpha. Oncogene 2006;25:3589-97.
3. Park J, Kim M, Lim J, Kim Y, Han K, Kim JS, et al. Variant of ETV6/ABL1 gene is associated with leukemia phenotype. Acta Haematol 2013;129:78-82.
4. Million RP, Harakawa N, Roumiantsev S, Varticovski L, Van Etten RA. A direct binding site for Grb2 contributes to transformation and leukemogenesis by the Tel-Abl (ETV6-Abl) tyrosine kinase. Mol Cell Biol 2004;24:4685-95.
5. Gancheva K, Virchis A, Howard-Reeves J, Cross NC, Brazma D, Grace C, et al. Myeloproliferative neoplasm with ETV6-ABL1 fusion: a case report and literature review. Mol Cytogenet 2013;6:39.
6. Song JS, Shin SY, Lee ST, Kim HJ, Kim SH. A cryptic ETV6/ABL1 rearrangement represents a unique fluorescence in situ hybridization signal pattern in a patient with B acute lymphoblastic leukemia. Ann Lab Med 2014;34:475-7.
7. Keung YK, Beaty M, Steward W, Jackle B, Pettnati M. Chronic myelocytic leukemia with eosinophilia, t(9;12)(q34;p13), and ETV6-ABL gene rearrangement: case report and review of the literature. Cancer Genet Cytogenet 2002;138:139-42.
8. De Braekeleer E, Douet-Guilbert N, Rowe D, Bown N, Morel F, Berthou C, et al. ABL1 fusion genes in hematological malignancies: a review. Eur J Haematol 2011;86:361-71.
9. La Starza R, Trubia M, Testoni N, Ottaviani E, Belloni E, Crescenzi B, et al. Clonal eosinophils are a morphologic hallmark of ETV6/ABL1 positive acute myeloid leukemia. Haematologica 2002;87:789-94.
10. Perna F, Abdel-Wahab O, Levine RL, Jhanwar SC, Imada K, Nimer SD. ETV6-ABL1-positive "chronic myeloid leukemia": clinical and molecular response to tyrosine kinase inhibition. Haematologica 2011;96:342-3.

https://doi.org/10.3343/alm.2017.37.1.77