Case Report

Molecular Monitoring in Adult Philadelphia Chromosome-Positive Acute Lymphoblastic Leukemia with the Variant e13a3 BCR-ABL1 Fusion

Mireille Crampe,1 Laura Kearney,1 David O’Brien,2 C. Larry Bacon,2 Derville O’Shea,3 and Stephen E. Langabeer1

1Cancer Molecular Diagnostics, St. James’s Hospital, Dublin 8, Ireland
2Department of Haematology, St. James’s Hospital, Dublin 8, Ireland
3Department of Haematology, Cork University Hospital, Cork, Ireland

Correspondence should be addressed to Stephen E. Langabeer; slangabeer@stjames.ie

Received 3 April 2019; Accepted 13 May 2019; Published 27 June 2019

1. Introduction

Philadelphia chromosome-positive acute lymphoblastic leukemia (Ph+ ALL) in adults is an aggressive disease that responds poorly to conventional chemotherapy. Despite improvements in survival with the addition of tyrosine kinase inhibitors (TKI) to chemotherapy, hematopoietic allogeneic stem cell transplantation (ASCT) remains the only curative option in those eligible patients [1]. Molecular monitoring of BCR-ABL1 transcripts is a valuable tool in assessing individual patient response to chemotherapy and ASCT [2–4]. The most common BCR-ABL1 transcripts in Ph+ ALL are the e1a2, e13a2, and e14a2 fusions [5]; however, approximately 5% of adult patients express variant BCR-ABL1 transcripts [6]. Of these variants, the e13a3 (b2a3) BCR-ABL1 type is extremely rare with scant information regarding optimal therapeutic approach [7, 8]. Characterisation of these rare BCR-ABL1 variants also affords the selection of appropriate primer/probe combinations for reverse-transcriptase quantitative PCR (RT-qPCR) assessment of residual disease. The presentation and clinical course of a patient with e13a3 BCR-ABL1 Ph+ ALL is reported.

2. Case Report

A 23-year-old male presented with chest pain and dyspnoea, a hemoglobin count of 7.4 g/dL, platelet count of <10 × 10⁹/L, and white cell count of 11.6 × 10⁹/L. Bone marrow (BM) biopsy and aspirate demonstrated a 95% infiltration of lymphoblasts (Figure 1). Immunophenotyping of the BM aspirate showed lymphoblasts were CD10-, CD19-, CD20-, CD34-, TdT-, and HLA-DR-positive. Cytogenetic analysis revealed a complex clone in 10 cells analysed by G-banding...
containing various structural and numerical abnormalities including a derivative chromosome 22 from a translocation between the long arms of chromosomes 9 and 22. The composite karyotype was 44–48, XY, +X, t(1; 14)(p32; q32), −3, −6, add(6)(p21), add(7)(p21), add(8)(p21), −10, add(12)(p13), +16, add(19)(p13), der(22)t(9; 22)(q34; q11.2), +1−2mar, inc [cp10]. Interphase FISH analysis showed the presence of BCR-ABL1 rearrangement in 64/100 cells analysed. Standardised RT-PCR and Sanger sequencing demonstrated e13a3 BCR-ABL1 transcripts [9] (Figure 2). E13a3 BCR-ABL1 transcripts lack ABL1 exon a2, thus prohibiting the use of a standardised primer/probe combination for e14a2/e14a2 BCR-ABL1 qPCR [10]. A modified BCR-ABL1 qPCR assay was therefore adopted utilising BCR forward primer ENF501F2 [9] with ABL1 reverse primer ENR1063 and ABL1 probe ENP1043, the latter both complementary to ABL1 exon a3 sequence [11]. The BCR-ABL1 standard curve was constructed using serial dilutions of an e14a3 BCR-ABL1 plasmid over a five log range (100% to 0.001% BCR-ABL1 transcripts) with ABL1 as the reference gene as previously described [12]. Best practice guidelines for BCR-ABL1 qPCR and data interpretation were followed [13]. Presentation e13a3 BCR-ABL1 transcripts were of a high level (BCR-ABL1/ABL1 75.7%) consistent with a diagnosis of pre-pre-B cell Ph+ ALL.

The patient commenced induction treatment with rituximab, dexamethasone, vincristine, and daunorubicin, with imatinib 400 mg oral daily started on day 15. After phase-one induction, the bone marrow aspirate demonstrated a complete morphological response with residual disease detected by immunophenotyping and RT-qPCR (BCR-ABL1/ABL1 8.11%). After phase-two induction, the bone marrow BCR-ABL1/ABL1 level had fallen to 0.053%. Following high-dose methotrexate, the pre-ASCT BCR-ABL1 level was 0.034%. The patient proceeded to ASCT from an unrelated donor after cyclophosphamide and total body irradiation conditioning and recommenced continued imatinib maintenance. BCR-ABL1 transcripts were not detected in the peripheral blood at one, two, and three and a half months post-ASCT (Figure 3). Continued close molecular monitoring is planned.

3. Discussion

E13a3 BCR-ABL1 transcripts lack ABL1 exon a2 that encodes part of SH3 domain thought to contribute to leukemiaogenesis by inhibition of the kinase domain and by STAT5 activation [6]. In chronic myeloid leukemia patients, this transcript results in an indolent and TKI-responsive form of disease [14–16]; however, its prognostic significance in adult Ph+ ALL patients remains unknown due to the limited number of annotated cases. While the possibility exists of lymphoid blast crisis in chronic myeloid leukemia (CML), this transformation in e13a3 BCR-ABL1 CML is rare [17]. In the absence of basophilia, thrombocytosis, and splenomegaly, this case likely represents de novo Ph+ ALL. Additional chromosomal abnormalities and complex karyotypes are frequently observed in Ph+ ALL, as witnessed in this case. There is some suggestion that in Ph+ ALL, additional cytogenetic abnormalities are associated with a shorter overall survival and might therefore be used for

Figure 1: Bone marrow morphology at diagnosis showing infiltration by lymphoblasts.

Figure 2: Sanger sequencing demonstrating presence of the e13a3 BCR-ABL1 fusion gene.
stratification purposes [18]. In the post-ASCT setting, monitoring BCR-ABL1 transcript levels is an essential component of Ph+ ALL patient management [19] with effective standardisation of RT-qPCR assays for this and other variant BCR-ABL1 fusion transcripts required [20].

Reporting of further cases would enable identification of any phenotypic characteristics of e13a3 BCR-ABL1 Ph+ ALL and help establishing optimal treatment strategies for patients with this rare genotype.

Conflicts of Interest

The authors declare that there are no conflicts of interest regarding the publication of this paper.

References

[1] M. Yilmaz, H. Kantarjian, F. Ravandi-Kashani, N. J. Short, and E. Jabbour, “Philadelphia chromosome-positive acute lymphoblastic leukemia in adults: current treatments and future perspectives,” Clinical Advances in Hematology & Oncology, vol. 16, no. 3, pp. 216–223, 2018.

[2] M. Yanada, I. Sugiura, J. Takeuchi et al., “Prospective monitoring of BCR-ABL1 transcript levels in patients with Philadelphia chromosome-positive acute lymphoblastic leukaemia undergoing imatinib-combined chemotherapy,” British Journal of Haematology, vol. 143, no. 4, pp. 503–510, 2008.

[3] L. Zhang, R. T. Ramjit, C. E. Hill, M. Arellano, H. J. Khouy, and K. P. Mann, “Clinical significance of quantitative monitoring and mutational analysis of BCR-ABL1 transcript in Philadelphia chromosome positive B lymphoblastic leukemia,” Leukemia & Lymphoma, vol. 57, no. 2, pp. 364–369, 2016.

[4] G. Cazzaniga, P. De Lorenzo, J. Alten et al., “Predictive value of minimal residual disease in Philadelphia- and chromosome-positive acute lymphoblastic leukemia treated with imatinib in the European intergroup study of post-induction treatment of Philadelphia- and chromosome-positive acute lymphoblastic leukemia, based on immunoglobulin/T-cell receptor and BCR/ABL1 methodologies,” Haematologica, vol. 103, no. 1, pp. 107–115, 2018.

[5] S. E. Langabeer, “Variant BCR-ABL1 fusion genes in adult Philadelphia chromosome-positive B-cell acute lymphoblastic leukemia,” EXCLI Journal, vol. 16, pp. 1144–1147, 2017.

[6] A. Tusznyski, S. Dhut, B. D. Young et al., “Detection and significance of bcr-ABL1 mRNA transcripts and fusion proteins in Philadelphia-positive adult acute lymphoblastic leukemia,” Leukemia, vol. 7, no. 10, pp. 1504–1508, 1993.

[7] T. Burmeister, S. Schwartz, A. Taubald et al., “Atypical BCR-ABL mRNA transcripts in adult acute lymphoblastic leukemia,” Haematologica, vol. 92, no. 12, pp. 1699–1702, 2007.

[8] X. Zhang and J. Pan, “An e13a3 BCR-ABL1 fusion transcript in variant t(9;22;17)(q34;q11;q21)-positive adult acute lymphoblastic leukemia,” International Journal of Laboratory Hematology, vol. 38, no. 3, pp. e52–e55, 2016.

[9] J. J. M van Dongen, E. A. Macintyre, J. A. Gabert et al., “Standardized RT-PCR analysis of fusion gene transcripts from chromosome aberrations in acute leukemia for detection of minimal residual disease,” Leukemia, vol. 13, no. 12, pp. 1901–1928, 1999.

[10] J. Gabert, E. Beillard, V. H. J. van der Velden et al., “Standardization and quality control studies of ‘real-time’ quantitative reverse transcriptase polymerase chain reaction chain reaction of fusion gene transcripts for residual disease detection in leukemia—a Europe against cancer program,” Leukemia, vol. 17, no. 12, pp. 2318–2357, 2003.

[11] E. Beillard, N. Pallisgaard, V. H. J. van der Velden et al., “Evaluation of candidate control genes for diagnosis and residual disease detection in leukemic patients using ‘real-time’ quantitative reverse-transcriptase polymerase chain reaction (RQ-PCR)—a Europe against cancer program,” Leukemia, vol. 17, no. 12, pp. 2474–2486, 2003.

[12] S. L. McCarron, S. E. Langabeer, K. Bolger et al., “Molecular response to imatinib in chronic myeloid leukemia with a variant e13a3 BCR–ABL1 fusion,” Medical Oncology, vol. 32, no. 2, p. 452, 2015.
[13] L. Foroni, G. Wilson, G. Gerrard et al., “Guidelines for the measurement of BCR-ABL1 transcripts in chronic myeloid leukaemia,” *British Journal of Haematology*, vol. 153, no. 2, pp. 179–190, 2011.

[14] D. S. Snyder, R. McMahon, S. R. Cohen, and M. L. Slovak, “Chronic myeloid leukemia with an e13a3 BCR-ABL fusion: benign course responsive to imatinib with an RT-PCR advisory,” *American Journal of Hematology*, vol. 75, no. 2, pp. 92–95, 2004.

[15] B. Pienkowska-Grela, R. Woroniecka, I. Solarska et al., “Complete cytogenetic and molecular response after imatinib treatment for chronic myeloid leukemia in a patient with atypical karyotype and BCR-ABL b2a3 transcript,” *Cancer Genetics and Cytogenetics*, vol. 174, no. 2, pp. 111–115, 2007.

[16] B. Liu, W. Zhang, and H. Ma, “Complete cytogenetic response to Nilotinib in a chronic myeloid leukemia case with a rare e13a3(b2a3) BCR-ABL fusion transcript: a case report,” *Molecular Medicine Reports*, vol. 13, no. 3, pp. 2635–2638, 2016.

[17] J. Ha, J.-W. Cheong, S. Shin, S.-T. Lee, and J. R. Choi, “Chronic myeloid leukemia with rare variant b2a3 (e13a3) BCR-ABL fusion,” *Annals of Laboratory Medicine*, vol. 36, no. 3, pp. 287–289, 2016.

[18] C. A. Seol, Y.-U. Cho, S. Jang et al., “Prognostic significance of recurrent additional chromosomal abnormalities in adult patients with Philadelphia chromosome-positive acute lymphoblastic leukemia,” *Cancer Genetics*, vol. 216-217, pp. 29–36, 2017.

[19] H. Pfeifer, B. Wassmann, W. Bethge et al., “Randomized comparison of prophylactic and minimal residual disease-triggered imatinib after allogeneic stem cell transplantation for BCR-ABL1-positive acute lymphoblastic leukemia,” *Leukemia*, vol. 27, no. 6, pp. 1254–1262, 2013.

[20] S. E. Langabeer, “Standardized molecular monitoring for Variant BCR-ABL1 Transcripts in chronic myeloid leukemia,” *Archives of Pathology & Laboratory Medicine*, vol. 139, no. 8, p. 969, 2015.