Double-Hit Lymphoma with t(8;14)(q24;q32) and t(12;14)(q24;q32) Chromosomal Translocations

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Abstract

A 50-year-old man presented with an ileocecal tumor and a large amount of ascites. Lymphoma cells obtained from the ascitic fluid were CD10⁺, CD20⁺, CD38⁺, HLA-DR⁺, BCL6, MUM1/IRF4⁺, BCL2⁺, and immunoglobulin μ/γ⁺. The karyotype determined by G-banding and spectral karyotyping was 46, XY, der(3) t(1;3)(q12;p12), -4, +7, t(8;14)(q24;q32), t(12;14)(q24;q32), der(17)t(4;17)(q21;p11). Fluorescence in situ hybridization disclosed that 93% of interphase cells were positive for the c-MYC and immunoglobulin heavy chain gene fusion. The patient was treated with intensive chemo-immunotherapy, resulting in a complete response. The t(8;14)-t(12;14) double-hit may have generated molecular abnormalities analogous to those of a previously cloned three-way translocation t(8;12;14).

Key words: double-hit lymphoma, spectral karyotyping, t(8;14)(q24;q32), t(12;14)(q24;q32)

Introduction

The 2008 WHO classification of hematolymphoid tumors lists the category of B-cell lymphoma, unclassifiable (BCL-U), with features intermediate between diffuse large B-cell lymphoma (DLBCL) and Burkitt lymphoma (BL) (1). Genetic abnormalities of this intermediate category included 8q24/c-MYC translocation involving either the immunoglobulin (Ig) genes or non-Ig genes as partners in combination with 18q21/BCL2 and/or 3q27/BCL6 translocation, and lymphomas with these double/triple translocations are referred to as “double-hit” or “triple-hit” lymphomas (1, 2). The clinical features of double/triple-hit lymphomas are aggressive, probably reflecting the deregulated expression of both proliferative (MYC) and anti-apoptotic (BCL2) proteins (2), while the synergistic mechanisms of transcriptional repressor BCL6 with MYC and/or BCL2 remains a subject to be clarified (3).

Here, we describe the case of an aggressive B-cell lymphoma that meets the criteria of BCL-U. Conventional cytogenetic analysis in addition to spectral karyotyping (SKY) revealed that lymphoma cells carried both t(8;14)(q24;q32) and t(12;14)(q24;q32).
Fisher E 1. Appearance of lymphoma cells contained in the ascitic fluids. Top, Giemsa-stained smear slide (original magnification, ×100 objective). Lymphoma cells around macrophages (A), mitotic figures (arrows in B), and an apoptotic cell with nuclear fragmentation (arrowhead in C) are shown. Bottom, immunohistochemistry of pathological specimens prepared from ascitic cells (original magnification, ×100 objective). The specimens were stained by anti-CD10 (D), anti-BCL6 (E), anti-MUM1/IRF4 (F), and anti-BCL2 (G) antibodies.

diagnosis of high-grade B-cell lymphoma, and treated the patient with rituximab plus cyclophosphamide, doxorubicin, vincristine, and prednisone (R-CHOP), followed by rituximab plus hyper-fractionated cyclophosphamide, vincristine, doxorubicin, and dexamethasone (hyper-CVAD), and rituximab plus high-dose methotrexate and cytarabine (4). As a response to the treatment, ascites resolved smoothly and the patient’s condition improved. After the completion of 4 cycles of the latter chemo-immunotherapy regimen, the patient achieved a complete remission.

Cytogenetic studies

G-banded karyotyping of metaphase spreads prepared form the ascitic cells readily revealed t(8;14)(q24;q32); however, small chromosomal materials seemed to be added to the end of the other chromosome 14 homologue (Fig. 2). Fluorescence in situ hybridization (FISH) using the c-MYC and Ig heavy chain gene (IgH) probe set showed that 93% of interphase cells were positive for the c-MYC-IgH gene fusion, while the majority of positive cells carried two IgH signals (Fig. 3A, left). The BCL2-IgH FISH was negative for the fusion, while the cells showed three or four IgH signals (Fig. 3B). These FISH results suggested the presence of additional translocation involving the other IgH locus. We therefore performed SKY to reveal the second 14q32/IgH translocation. As shown in Fig. 4, t(8;14)(q24;q32) was confirmed and additional materials of the other der(14) were from the long arm of chromosome 12 with the breakpoint at 12q24 and 14q32. Trisomy 1q and 17p abnormality became clearer by SKY. The karyotype determined by the G-banding and SKY was: 46, XY, der(3)t(1;3)(q12;p12), -4, +7, t(8;14)(q24;q32), t(12;14)(q24;q32), der(17)t(4;17)(q21;p11).

Discussion

Double-hit lymphomas other than the combination of c-MYC with BCL2 and/or BCL6 have been reported in the literature, including 8q24/c-MYC translocation with 11q13/CCND1, with 19q13/BCL3, and with 9p13/PAX5 translocations (2). We showed here that the lymphoma carried t(8;14)(q24;q32)/c-MYC-IgH gene fusion in combination with t(12;14)(q24;q32) involving the other IgH locus. As we observed interphase cells showing one c-MYC, one IgH, and two c-MYC-IgH fusion signals (Fig. 3A, right), the t(12;14) is considered to be the second hit following the t(8;14). The
12q24 locus affected by t(12;14) at the cytogenetic level contains the BCL7A gene, which was isolated by cloning a three-way translocation t(8;12;14)(q24.1;q24.1;q32.3) in a BL cell line, Wien 133 (5, 6). The gene encodes a protein showing homology with the actin-binding protein caldesmon (6). As the result of t(8;12;14), exon I of BCL7A was replaced by c-MYC exon I, leading to the production of chimeric transcripts of c-MYC-BCL7A; however, the BCL7A mRNA level in Wien 133 seemed to be low compared with controls (6). Although it is possible that the t(8;14)-t(12;14) double-hit of the present case generated molecular abnormalities analogous to those of t(8;12;14), to determine whether the t(12;14) targeted the BCL7A and whether the gene was fused with IgH instead of c-MYC, cloning and sequencing of the t(12;14) breakpoint are required.

Clinical features of the present case shared those of classical BL, in that the disease initially presented with an ileocecal tumor and large ascites suggesting a high tumor burden (7). The morphology of lymphoma cells apparently resembled that of BL cells. On the other hand, expression of BCL2 confirmed by both immunohistochemistry and flow cytometry and expression of MUM1/IRF4 as well as complex cytogenetic abnormalities other than t(8;14) may not favor the diagnosis of BL (1, 7, 8). We therefore conclude that the lymphoma of this case does not exactly meet the criteria of BL but can be placed into the BCL-U category of the WHO 2008 classification (1, 2).

Treatment of double-hit lymphoma has not been established. Patients have been variably treated with conventional chemotherapy regimens for DLBCL, intensive chemotherapy regimens developed for childhood acute lymphoblastic leukemia, and high-dose chemotherapy with autologous hematopoietic stem cell support (9-11). Niitsu et al suggest that patients, in general, show a response to treatment, but finally suffer early relapse, resulting in poor progression-free and overall survival rates (10). Nevertheless, it is currently unknown whether the long-term outcome of this particular case with t(8;14)-t(12;14) double hit is comparable to those of c-MYC-BCL2 double-hit and c-MYC-BCL2-BCL6 triple-hit lymphomas.
The authors state that they have no Conflict of Interest (COI).

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References

Figure 4. Spectral karyotyping (SKY). G-banded chromosomes and their pseudo-color images by SKY are aligned side by side. Bottom, enlarged images of chromosomes 8, 14, and 12. Closed and open arrows indicate t(8;14)(q24;q32) and t(12;14)(q24;q32), respectively.