B-cell acute lymphoblastic leukemia with t(4;11)(q21;q23) in a young woman: evolution into mixed phenotype acute leukemia with additional chromosomal aberrations in the course of therapy

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Abstract

About 5% of adult B-cell acute lymphoblastic leukemias (B-ALL) are characterized by t(4;11)(q21;q23), which confers peculiar features to this B-ALL subtype, including a very immature immunophenotype and poor prognosis. We describe the case of a 21-year-old female who presented with B-ALL carrying the t(4;11)(q21;q23) and blasts positive for CD19, TdT, CD79a, CD33, CD38, CD58, HLA-DR. Before completing the Hyper-CVAD (hyperfractionated cyclophosphamide, vincristine, doxorubicin, and dexamethasone) therapy regimen, an additional leukemic clone appeared, with morphology and immunophenotypic features, appeared in the course of induction therapy, probably accelerating the fatal outcome of the patient. Cyto genetics and in situ fluorescent hybridization (FISH) showed the co-existence of t(4;11)(q21;q23) with a complex karyotype, which was characterized by three trisomies and the presence of two deriva- tives of chromosome.

Therefore, the initial B-ALL with t(11;14) (q21;q23) showed evolution into a bilinear lineage acute leukemia (lymphoid and myeloid) compatible with the 2008 WHO entity defined as mixed phenotype acute leukemia (MPAL) with t(11;14), MLL rearranged. To the best of our knowledge, a similar evolution of B-ALL with t(11;14) (q21;q23) has not been described so far.

Case Report

A Caucasian 21-year-old female presented at the Division of Hematology of the University of Pisa, Italy, with fever and anemia-related symptoms. A complete blood count showed hyperleukocytosis [white blood cell (WBC) 400 x 10^9/L], anemia and thrombocytopenia (9 g/dL and 50 x 10^9/L, respectively). Her clinical history was silent, but she declared intake of heroin and cocaine. Physical examination showed mild splenomegaly. Whole body computed tomography confirmed the spleen enlargement (15 cm longitudinal diameter) and did not show pathologic lymph nodes.

Manual WBC differential count of peripheral blood showed 90% blasts without morphologic differentiation (Figure 1A), 2% neutrophils, 8% small lymphocytes. Blasts resulted negative for myeloperoxidase stain. Flow cytometric analysis was therefore performed using a wide monoclonal antibody panel and a six-color method: blasts were positive for CD19, TdT, CD79a, CD33, CD38, CD58, HLA-DR (Figure 2A-F).

Bone marrow samples obtained from an aspirate were processed for light microscope evaluation, immunophenotyping, cytogenetics, polymerase chain reaction (PCR) assays for IgH gene rearrangement.

A massive infiltration by blasts with apparent lymphoid morphology was detected and immunophenotyping confirmed the results obtained with peripheral blood evaluation.

To perform PCR assays for IgH gene clonality assessment, monoclonal cells were separated by Ficoll/Hypaque gradient from bone marrow, and suitable aliquots were utilized for PCR tests after spectrophotometric quantitative evaluation. Fluorescent PCR reactions for IgH clonality evaluation were carried out with CDR3-specific consensus primer and analyzed by ABI PRISM 3100 (Applied Biosystems). PCR tests showed a clonal IgH gene rearrangement. Finally cytogenetics, performed by using the Q-banding technique, revealed a karyotype with t(4;11)(q21;q23) in 90% of metaphases.

The final diagnosis B-ALL with t(4;11)
Chemotherapy with the dose-intensive phase of hyperfractionated cyclophosphamide, vincristine, doxorubicin, and dexamethasone (Hyper-CVAD) regimen was therefore given. Therapy included 8 cycles of dose-intensive therapy courses of Hyper-CVAD (courses 1, 3, 5, and 7) alternating with high-dose methotrexate and cytosine arabinoside (courses 2, 4, 6, and 8). Intrathecal central nervous system prophylaxis was also given with methotrexate or cytosine arabinoside, and recombinant human granulocyte colony-stimulating factor was administered as a supportive care.

After six of the established eight courses of therapy, although complete clearance of blasts from peripheral blood was obtained, minimal residual disease (0.2% by flow cytometry), was detected in the bone marrow, along with the presence of a small leukemic clone in cerebrospinal fluid samples. The final two cycles of therapy were programmed, but the patient refused further antiblastic therapies and was dismissed from our hospital.

At home, her clinical conditions showed a rapid worsening and two weeks later she was admitted at Versilia Hospital, Italy. The patient presented with hyperleukocytosis (220 x 10^9/L), hemoglobin 7 g/dL, platelets 10 x 10^9/L, cutaneous hemorrhages (petechiae, purpura), laboratory findings of disseminated intravascular coagulation (low fibrinogen, prolonged prothrombin time and a-PTT, high D-dimer levels, low antithrombin levels), fever.

The observation of peripheral blood smears showed blasts 95%, which consisted of two different clones, the former being represented by blast cells with lymphoid appearance (about 10%), the latter being represented by cells with a larger size, abundant cytoplasm, giant nucleus with irregular profile (about 90%) (Figures 1B and 1C). Some cells were classified as atypical monocytoid cells (arrowheads) and very few blasts showed cytoplasmic granulations (Figure 1B). Immunophenotyping of circulating blasts showed a peculiar CD45/SSC dot-plot, with the presence of two distinct blast cell populations. Blasts with small forward scatter (FSC) and side scatter (SSC), which accounted for 10%, showed the phenotypic characteristics observed at diagnosis and were classified as belonging to the B-cell lineage. The majority of blasts (90%) were characterized by larger FSC and SSC and appeared to derive from a myeloid/monocytic clone, being positive for CD13, CD33, CD64, CD15, CD56 and CD4 dim (Figure 3).

A new evaluation of karyotype confirmed the presence of the sole abnormality t(4;11)(q21;q23) in 10% of metaphases, and showed the appearance of two additional cell lines: one with 50 chromosomes, t(4;11)(q21;q23), trisomy of chromosomes number 8, 12 and 13, and a derivative of chromosome 4 [der(4)] (Figure 4B). This latter anomaly was confirmed by FISH, which was performed on metaphases using whole chromosome painting n. 4 and n. 11 probes (Cytocell Inc. UK) and detected two der(4) (Figure 5). It was not possible to perform other studies, with the exception of a new PCR for IgH rearrangement, which showed persistence of a clonal pattern. The patient died because of disseminated intravascular coagulation.

**Discussion**

The B-ALL subtype carrying t(4;11)(q21;q23) is a rare event, representing 5-10% of adult
cases of B-ALL, which, in turn, is a disease with very low incidence (less than 1 per 100,000 persons/year). In an ample study published by the Medical Research Council and the Eastern Cooperative Oncology Group, which involved 1522 patients with acute lymphoblastic leukemia, only 54 showed a B-ALL with t(4;11)(q21;q23).\(^9\) This type of B-ALL shows peculiar features. From a clinical point of view, it is characterized by elevated WBC counts, high incidence of central nervous system involvement, frequent hepatosplenomegaly, poor clinical outcome both in children and in adults.\(^6\) Due to these reasons, B-ALL with t(4;11) are considered as high-risk leukemias.\(^5\) The presence of t(4;11)(q21.q23) is more frequent in B-ALL deriving from a very immature B-cell precursor.\(^5\) This feature is suggested by the phenotype displayed by blast cells, which are generally positive for CD19 and markers associated with an immature immunophenotype, as demonstrated by the co-expression of CD19, TdT, CD79a and CD34, and the absence of CD10 and CD20. Negativity for CD10 is always observed.\(^10\) Because of this peculiar immunophenotype, the B-ALL subtype carrying t(4;11)(q21;q23) represents about 40% of all forms of pro-B ALL in adults. Several but not all cases show co-expression of some myeloid markers, such as CD15 and CD65, with constant negativity of CD13 and CD33.\(^4\)

In our case findings of typical B-ALL with t(4;11)(q21;q23) were found at diagnosis. Central nervous system involvement was detected during the course of disease, in a phase of bone marrow minimal residual disease.

The poor outcome of our patient was favored by an unexpected event, represented by the appearance of an additional leukemic clone with both morphological and immunophenotypical properties which could be attributed to the myeloid/monocytic lineage. In fact, during the terminal and fatal phase of disease, two distinct blast cell populations were found, the former with a B-cell phenotype and the latter with a separate phenotype characterized by co-expression of markers frequently found in acute myeloid leukemias with prevalent monocytic differentiation.\(^11,12\) Such as CD13, CD33, CD64, CD15, CD4. Co-expression of B-cell markers and myeloid markers was not detected. The appearance of this additional leukemic clone was accompanied by the preponderant presence of additional cytogenetical aberrations, such as trisomy of chromosomes 8, 12 and 13. Trisomy 8 is a relatively frequent (10-15% of cases) abnormality in acute myeloblastic leukemias (AML), in which is associated with a poor prognosis. Isolated trisomy 12 is an infrequent finding both in AML and in B-ALL and has been described in highly undifferentiated acute leukemias. Isolated trisomy 13 is another marker with a negative impact on prognosis and seems to be associated with the FAB M0 subtype and very low remission rates. Thus, the association of these three cytogenetical anomalies should be interpreted as biologic marker with an additional negative impact on disease.

The presence of double der(4) [derivative of t(4;11)(q21;q23)] is likely to have given a synergistic effect to the leukemic phenotype as well as an attractive leukemic subtype with poor prognosis. The latter finding is consistent with the initial lymphoblastic clone, since a der(4) chromosome can be found in up to 65% of patients with t(4;11)(q21;q23).\(^15\)

There is evidence of an interesting association of MLL gene rearrangements and MPAL. The 2008 WHO classification has identified a particular subtype of MPAL termed mixed phenotype acute leukemia with t(11;qlq23); MLL rearranged,\(^3\) which can show either the presence of blasts with simultaneous expression of markers of different lineages or the presence of two populations of blasts with distinct phenotype. In a recent analysis of 100 cases of MPAL, Matutes et al.\(^14\) found only three cases with MLL gene rearrangement due to t(4;11)(q21;q23) at initial presentation. In a less recent report Johansson et al.\(^15\) analyzed 183 cases of t(4;11)(q21;q23) and found that 34% were children, 95% were B-ALL and that only one case was a biphenotypic acute leukemia. Rubnitz et al.\(^16\) reported a series of 35 children with MPAL and found four cases with MLL gene rearrangement: only two cases showed a bilineal pattern (myeloid and lymphoid B), but t(4;11)(q21;q23) was not detect-
ed. Sporadic additional cases of acute biphenotypic leukemia associated with complex MLL gene rearrangement have been reported.17,18

Thus, although MLL gene rearrangement with t(4;11)(q21;q23) can be observed in cases of MPAL, this peculiar subtype of acute leukemia seems to be very rare at first presentation, probably being more frequent in children.

Another possible presentation of cases of B-ALL with t(4;11)(q21;q23) is that leukemic opinion about the existence of a common lymphoid-myeloid precursor,23 is that leukemic blasts of myeloid lineage following expansion of a pre-existing minor population that the lineage switch might represent the rare case of B-ALL with t(4;11)(q21;q23). Not only phenotypic switches are possible evolution of B-ALL with t(4;11) and BPAL, early detection of such uncommon evolution of this B-ALL sub-type may have practical implications in terms of follow-up strategies.

Conclusions

Because of the above considerations, it appears that in our patient the initial B-ALL with t(4;11)(q21;q23) showed, in the course of therapy, an evolution into a MPAL with the same MLL gene rearrangement, but with additional chromosomal aberrations probably conferred by the additional, non-lymphoblastic, leukemic clone.

We think that the case described in the current report adds new information about the possible evolution of B-ALL with t(4;11)(q21;q23). Not only phenotypic switches are possible after chemotherapy, but additional leukemic clones, with myeloid/monocytic properties, may arise in the course of antiblastic therapy, probably deriving from a common precursor. Thus, a wide panel of MoAb should be used when monitoring B-ALL positive for t(4;11)(q21;q23), in order to detect eventual phenotypic shifts and/or appearance of additional leukemic clones. Due to the poor outcome which characterizes both B-ALL with t(4;11)(q21;q23) and MPAL, early detection of such uncommon evolution of this B-ALL sub-type may have practical implications in terms of follow-up strategies.

References


