In most cases a differential diagnosis between different acute leukemias, lymphoid and myeloid, can be readily established by morphological examination and cytochemical staining. Nevertheless, in a portion of cases, immunophenotype analysis has revealed the coexpression of lymphoid and myeloid antigens in the same cells. Terms such as biphenotypic, mixed-lineage or hybrid leukemias have been used in these cases.

Several reports have analyzed myeloid antigen expression in acute lymphoblastic leukemias; however, data on their frequency and clinical significance are conflicting, probably due to lack of standardization in the definition myeloid antigen expression, immunophenotyping techniques and treatment. Translocation t(8;14) and the variants t(8;22) and t(2;8) has been considered characteristic of high-grade Burkitt’s and non-Burkitt’s small non-cleaved cell type lymphomas, as well as of Burkitt’s type B-cell acute lymphoblastic leukemia (L3). Rare cases of lymphomas other than Burkitt’s with these translocations have also been reported. On the other hand, monosomy of chromosome #7 is the commonest numerical abnormality in acute non lymphocytic leukemias (ANLL).

In this paper we present a case of acute lymphoblastic leukemia (ALL) expressing myeloid antigens with a novel karyotypic association, t(8;22) and monosomy 7, and high Ag-NOR activity were found concomitantly with a very short survival.

**Case report**

A 6-year-old male patient was admitted to the hospital in June 1993. Clinical examination showed fever, cervical and retroauricular lymphadenopathies and hepatosplenomegaly. Hemogram at that date was: Hct 24%, WBC 550 × 10⁹/L, platelets 27 × 10⁹/L. LDH 1500 U/L (normal 250 U/L). The bone marrow (BM) showed 90% blast cells of variable size (8-20 μ) with occasional prominent nucleoli and cytoplasmic basophilia (Figure 1). Granulations, Auer rods and vacuoles were not observed. Blast...
cells in the BM were negative for the PAS reaction and peroxidase stain.

Immunophenotyping of BM mononuclear cells showed positivity for the following monoclonal antibodies: CD34 87%, HLA-DR 90%, CD10 26%, CD19 72%, Igs 5%, CD3 5%, CD7 4%, CD13 57% and CD33 47%. A diagnosis of ALL, FAB morphologic subtype L2, with myeloid markers was made.

The patient was treated with combination chemotherapy following a protocol like BFM (1-ALL-90 GATLA). Later he received intermediate methotrexate doses and high doses of cytosine arabinoside (Ara-C). He showed a poor response characterized by peripheral pancytopenia without medullar remission and immediate hematic invasion of leukemic blasts. He never achieved a complete remission and died in December 1993.

Materials and Methods

BM samples were processed for cytogenetic analysis by short-term (24 h) culture in F-10 medium with 15% fetal calf serum. Slides were prepared by a conventional method. A total of 30 cells were analyzed by the G-banding technique. The mean number of Ag-NOR chromosomes and the percentage of Ag-positive cells were studied by the Ag-NOR technique.

Fluorescence in situ hybridization (FISH) with a chromosome 7-specific α satellite DNA probe (SpectrumCEP chromosome 7, Spectrum Green, Imagenetics) for the centromeric region of this chromosome was performed according to the standard protocol (Imagenetics). The slides were counter-stained with propidium iodide. Hybridization signals were analyzed on the cytogenetic preparations from the patient and from a BM control in 440 interphase nuclei. A normal donor for BM transplantation was selected as control.

Results

Two abnormal clones with the following karyotypes were found: 45,XY,-7,t(8;22) (q24;q11) (62% of cells), and 46,XY,t(8;22) (q24;q11) (35% of cells) (Figure 2a). Three percent normal cells were also observed. The karyotype of the control was 46,XY. FISH analysis on preparations from the same sample (Figure 2b) showed that the percentage of monosomic cells was lower (31.3%) than that obtained by karyotypic analysis. Table 1 summarizes the cytogenetic and FISH results from the patient and the control. Ag-NOR staining showed a total of 6.83±0.91 Ag-NOR chromosomes per cell (mean±SD), and 96.4% Ag-positive mitoses. All the cells analyzed showed an Ag-positive 22q– marker chromosome.

Discussion

In this paper, we present an ALL patient with myeloid markers showing the cytogenetic association t(8;22) and monosomy 7, which to the best of our knowledge has not been described previously. Conflicting results have been reported with regard to the rates of remission and disease-free survival among patients with ALL and myeloid-antigen expression. A poor response and a very short survival were observed in our patient. A wide variety of cytogenetic abnormalities have been described in patients with myeloid antigen-positive ALL, but no cases with t(8;22) have been reported. Hanson referred a case of CD33+ ALL with monosomy 7 and other structural abnormalities. In our patient, the coexpression of lymphoid and myeloid markers and the presence of t(8;22) and monosomy 7 in the same cell may represent the malignant transformation of a pluripotent progenitor cell coexpressing features of both lineages.
When interphase FISH was compared to karyotype analysis for the detection of monosomy 7 leukemic cells, a lower percentage of cells were identified by the former technique. Similar results were observed by studying patients with ANLL and myelodysplastic syndrome. This discrepancy may reflect the proliferative advantage of the monosomic clone.

Both the percentage of Ag-NOR positive cells and the mean number of Ag-NOR chromosomes in our patient were higher than those observed in normal BM cells from healthy individuals. NOR activity in hemopoietic cells depends strongly upon the process of differentiation. Moreover, the area of interphase Ag-NORs is correlated with cell growth and is an important prognostic factor in childhood ALL. The high NOR activity found in this patient might reflect the degree of immaturity and the high proliferative activity of his bone marrow cells, which would be consistent with his negative clinical evolution. To date, the clinical significance of chromosome abnormalities in myeloid antigen-positive ALL is not clear. More studies will be necessary to clarify the role of cytogenetics as a prognostic factor in these pathologies.

Table 1. Comparison of karyotypic and FISH data.

<table>
<thead>
<tr>
<th>Karyotype</th>
<th>Nuclei with hybridization signal/s (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>0</td>
</tr>
<tr>
<td>Control</td>
<td>46,XY (100)</td>
</tr>
<tr>
<td>Patient</td>
<td>45,XY,-7,t(8;22)</td>
</tr>
</tbody>
</table>

References

10. Pedrazzini E, Slavutsky I. Ag-NOR staining and satellite association in bone marrow cells from patients with mycosis fungoides. Hereditas (In press).