Trisomy 8 in Acute Myeloblastic Leukemia and Sideroachrestic Anemia


An extra chromosome number 8 was found in the bone marrow of one patient with acute myeloblastic leukemia and one with sideroachrestic anemia. The extra chromosome was not found in the peripheral blood of any of the patients. In remission after treatment with cytotoxic drugs, the patient with acute myeloblastic leukemia had normal 46 XX cells in the bone marrow. In relapse, the same patient demonstrated two reciprocal translocations in addition to trisomy 8. This was interpreted as evidence for successive steps of clonal evolution from a single ancestor cell.

An extra C chromosome has been found in bone marrow cells from patients with a number of different malignant and premalignant blood disorders. To our knowledge, an identification of the extra chromosome by analysis of the chromosome banding pattern has so far only been reported in three patients with ineffective erythropoiesis, and all turned out to have trisomy 8. In the present paper, we report that trisomy 8 also occurs in acute myeloblastic leukemia. A further case with this chromosome abnormality in sideroachrestic anemia is also presented.

CASE REPORT

Case 1. Acute Myeloblastic Leukemia

The patient, a 55-yr-old woman, had previously had a gallstone attack in 1963. She was operated upon because of renal calculus in the left kidney in 1971. Since 1971, she had repeatedly had urinary tract infections.

Fourteen days before admission to the hospital in December 1972, she observed petechiae and ecchymosis on her legs and blood-stained expectoration. She also had anorexia and fatigue. On admission December 19, 1972, petechiae were found on the arms, legs, and buccal mucosa. Lymph glands and spleen were not enlarged. She had a moderate rise in temperature to 38.2°C. Hematologic investigations showed an acute myeloblastic leukemia (Table 1).

Treatment with the following drug combination was started immediately: Rubidomycine (Cerubidin, Pharma Rhodia), 1.5 mg/kg intravenously day 1, cytosine arabinoside (Cytosar, Upjohn), 1 mg/kg twice daily intravenously days 1-5, prednisolone (Precortalon Aquosum, Organon), 1 mg/kg twice daily intravenously days 1-5. This course of treatment was repeated three times with intervals of 5-28 days until remission was obtained. Remission was established...
<table>
<thead>
<tr>
<th>Sampling Occasion and No.</th>
<th>Hb (g/100 ml)</th>
<th>WBC (cells/µl)</th>
<th>Differential Counts (%)</th>
<th>Platelets (cells/µl)</th>
<th>Bone Marrow</th>
</tr>
</thead>
<tbody>
<tr>
<td>Patient No. 1 (AML)*</td>
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<tr>
<td>Dec 19, 1972 (1)</td>
<td>9.9</td>
<td>24.700</td>
<td>Myeloblasts 90.0</td>
<td>10.000</td>
<td>AML with 82% blast cells</td>
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<td></td>
<td>Promyelocytes 4.0</td>
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<td>Segments 2.0</td>
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<td></td>
<td></td>
<td></td>
<td>Lymphocytes 3.0</td>
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<td></td>
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<tr>
<td>Feb 7, 1973 (2)</td>
<td>9.8</td>
<td>3.500</td>
<td>Segments 42.5</td>
<td>353.000</td>
<td>Partial remission with 11% blast cells</td>
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<td></td>
<td></td>
<td></td>
<td>Bands 3.0</td>
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<td></td>
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<td>Lymphocytes 53.5</td>
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<td></td>
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<td>Monocytes 1.0</td>
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<tr>
<td>March 5, 1973 (3)</td>
<td>11.2</td>
<td>7.100</td>
<td>Metamyelocytes 0.5</td>
<td>590.000</td>
<td>Complete remission with 3% blast cells</td>
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<td></td>
<td></td>
<td></td>
<td>Segments 54.5</td>
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<td></td>
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<td></td>
<td>Bands 17.0</td>
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<td>Lymphocytes 18.5</td>
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<td></td>
<td></td>
<td>Monocytes 9.0</td>
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<tr>
<td>June 6, 1973 (4)</td>
<td>13.2</td>
<td>5.400</td>
<td>Myeloblasts 44.0</td>
<td>387.000</td>
<td>Relapse with 70% blast cells</td>
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<td></td>
<td></td>
<td>Segments 15.0</td>
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<td>Bands 3.0</td>
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<td></td>
<td></td>
<td>Lymphocytes 35.0</td>
<td></td>
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<td></td>
<td></td>
<td></td>
<td>Monocytes 3.0</td>
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<tr>
<td>Patient No. 2</td>
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<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>(Sideroblastic anemia)</td>
<td></td>
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<td></td>
</tr>
<tr>
<td>May 25, 1971</td>
<td>5.7</td>
<td>2.000</td>
<td>Segments 42.0</td>
<td>340.000</td>
<td>Hyperplastic megaloblastic erythroipoiesis. 88% sideroblasts of which 73% were ring sideroblasts. Abundant iron in reticulum cells.</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Bands 4.0</td>
<td></td>
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<td>Eosinophils 4.0</td>
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<td>Basophils 2.0</td>
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<td>Lymphocytes 43.0</td>
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<td></td>
<td></td>
<td>Monocytes 5.0</td>
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</tr>
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</table>

*AML, acute myeloblastic anemia.
in March 1973. A relapse occurred in June 1973 during reinduction therapy with cytosine arabinoside and thioguanine. Bone marrow aspiration and venous blood sampling for chromosome analyses were made on different occasions: (1) Dec 19, 1972, i.e., before treatment, (2) February 7, 1973, when there was a partial remission, (3) March 5, 1973, when there was a complete remission, and (4) June 6, 1973, when there was a relapse.

The hematologic data on the four sampling occasions are given in Table I.

Case 2. Sideroachrestic Anemia

The patient, a 69-yr-old woman, had previously been treated for pyelitis and thyroid dysfunction. A meningeoma had successfully been removed in 1963. An iron refractive anemia was diagnosed in 1969 and had been treated with blood transfusions.

She was admitted in 1970, and the laboratory investigations from one of her first visits to the hospital gave the following results: Hb 5.7 g/100 ml, RBC 1.9 million/cu mm, MCHC 35%, MCV 95 cu μ, reticulocytes 1.2%, serum iron 160 μg/ml, total iron binding capacity 300 μg/ml, serum vitamin B12 545 pg/ml, serum folic acid 1.5 ng/ml, LDH 290 U, sedimentation rate 30 mm/hr. White blood cell count, differential count, platelets, and bone marrow investigation are given in Table I. Liver function tests and routine urine analysis were normal. PBI and serum electrophoresis were also normal. Hemoglobin electrophoresis showed normal values, Hb A2 2%, and Hb F 1.2%.

Repeated bone marrow aspirates showed an active marrow with megaloblastoid traits. On the first sampling occasion for chromosome analysis on May 25, 1971 (Table I), 88% of the nucleated red cells were sideroblasts, 73%, of which were ring sideroblasts. The reticulum cells were heavily loaded with iron. On the second sampling occasion, 10 wk later, the hematologic picture was mainly the same.

The patient was treated with Pyridoxin, 80 mg orally three times daily from November 1970 to October 1972, i.e., also at the time of the chromosome analysis. She was later treated with Folacix, 5 mg three times daily orally, and from the middle of July to the end of 1971 with pyridoxal-5-phosphate. Although there has been no significant increase in the hemoglobin values, she is doing quite well and is living an apparently normal life.

MATERIALS AND METHODS

Chromosome analyses were made on air-dried cells from conventional leukocyte cultures after 72 hr of incubation using phytohemagglutinin, as well as on cells from bone marrow aspirates. In order to secure optimal conditions for a successful result, the bone marrow preparations were in each case made both directly without previous incubation and after 24 hr of incubation at 37°C without the use of phytohemagglutinin. Colchicine (0.125 μg/ml culture medium) was added 2 hr before harvesting. Fluorescence analysis of chromosomes was made according to the method of Caspersson et al. The nomenclature of the Paris Conference (1971) was used.

RESULTS

The results of the cytogenetic analysis are presented in Table 2.

Case I was investigated on four occasions. On the first occasion (before treatment), the direct preparation was successful, and only cells with 47 chromosomes and an extra chromosome number 8 were found. On the second occasion, the patient was in partial remission, and normal 46 XX as well as trisomic 47 XX + 8 cells were observed. On the third occasion, when the patient was in a complete hematological remission, only normal 46 XX cells were observed. On the fourth occasion, when there was a relapse, the cytogenetic picture was more complicated. A derivative of the original aneuploid cell line was then found in the bone marrow. All of 30 cells subjected to fluorescence analyses contained in addition to trisomy 8, two reciprocal translocations, one between chromosomes 2 and 18, and another one between chromosomes 9 and 15 (Fig. 1). The corresponding translocation
Table 2. Results of the Chromosome Analyses

<table>
<thead>
<tr>
<th>Case</th>
<th>Tissue</th>
<th>Date</th>
<th>Number of Cells</th>
<th>Number of Chromosomes</th>
<th>Karyotype</th>
</tr>
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<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>Bone marrow</td>
<td>Dec 19, 1972</td>
<td>25</td>
<td>25</td>
<td>47 XX +8</td>
</tr>
<tr>
<td></td>
<td>direct</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Bone marrow</td>
<td>Feb 7, 1973</td>
<td>11, 7</td>
<td>18</td>
<td>46 XX/47 XX +8</td>
</tr>
<tr>
<td></td>
<td>24 hr incubation</td>
<td></td>
<td></td>
<td></td>
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</tr>
<tr>
<td></td>
<td>Bone marrow</td>
<td>March 5, 1973</td>
<td>1, 14</td>
<td>15</td>
<td>46 XX</td>
</tr>
<tr>
<td></td>
<td>direct</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Bone marrow</td>
<td>June 6, 1973</td>
<td>55</td>
<td>55</td>
<td>47 XX +8, t(2;18)(p13;q23), t(9;15)(p11;q11)</td>
</tr>
<tr>
<td></td>
<td>direct</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Blood*</td>
<td>Dec 19, 1972</td>
<td>24</td>
<td>25</td>
<td>46 XX</td>
</tr>
<tr>
<td></td>
<td>Blood*</td>
<td>June 6, 1973</td>
<td>25</td>
<td>25</td>
<td>46 XX</td>
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<tr>
<td>2</td>
<td>Bone marrow</td>
<td>May 25, 1971</td>
<td>3, 4</td>
<td>8</td>
<td>46 XX/47 XX +8</td>
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<td></td>
<td>24 hr incubation</td>
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</tr>
<tr>
<td></td>
<td>Bone marrow</td>
<td>Aug 14, 1971</td>
<td>9, 16</td>
<td>25</td>
<td>46 XX/47 XX +8</td>
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<td></td>
<td>24 hr incubation</td>
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</tr>
<tr>
<td></td>
<td>Blood*</td>
<td>May 25, 1971</td>
<td>24</td>
<td>25</td>
<td>46 XX</td>
</tr>
</tbody>
</table>

*Seventy-two hours incubation in the presence of phytohemagglutinin.

...figures were also seen in 25 cells analyzed after conventional Giemsa staining. There were no other structural rearrangements or chromosome breaks.

Case 2 showed originally two cell lines in the bone marrow. One of the cell lines had a normal 46 XX and the other a 47 XX +8 karyotype (Fig. 2). At a second investigation a few months later, this picture remained unchanged.

No cells with an extra C chromosome were found in the leukocyte cultures from either of the two patients.

Fig. 1. Quinacrine fluorescent karyotype of bone marrow cell from patient with 47 XX +8, t(2;18)(p13;q23), t(9;15)(p11; q11) and acute myeloblastic leukemia.
DISCUSSION

Trisomy C in blood and bone marrow cells has been reported in a number of hematological diseases, namely, acute granulocytic leukemia, myelomonocytic leukemia, chronic myelocytic leukemia, erythroleukemia, polycythemia vera, and sideroachrestic anemia. It has previously been possible to identify the extra C chromosome as a number 8 in three cases of sideroachrestic anemia using fluorescence analysis. To our knowledge, the exact identification of the extra C chromosome present in some cases of acute myeloblastic leukemia has so far not been established. In the present report, it was possible to identify the extra C chromosome as a number 8 in one case of typical acute myeloblastic leukemia and one further case of sideroachrestic anemia.

Many patients with refractory sideroblastic anemia develop acute myelogenous leukemia or erythroleukemia, and chromosome abnormalities, including C trisomy, have been found in such cases. The present finding that the extra C chromosomes in these cases may be identical (No. 8) and appear both in preleukemic bone marrow, dominated by erythropoietic cells (sideroblasts), and in leukemic marrow, dominated by myelopoietic leukemic cells, supports the view of a common stem cell for erythropoietic and myelopoietic cells. Furthermore, these cells with trisomy 8 appear to have a selective advantage over the normal cell population in the bone marrow.

Only normal cells were found after treatment of the patient with acute myeloblastic leukemia with a combination of cytotoxic drugs. Such normalization in the bone marrow during remission has previously been demonstrated in different types of leukemia. The treatment of the present patient apparently suppressed the chromosomally abnormal cell population with persistence of a
more drug-resistant, cytologically and cytogenetically normal stem-cell population. This seems to be in contrast to the effect of the busulfan treatment of patients with chronic myelocytic leukemia. In most patients, clinical remission is not associated with the disappearance of the Ph¹ (No. 22)¹⁵ positive cell population in the bone marrow,¹⁶ even if a growing number of initially Ph¹ positive chronic myelocytic leukemia patients have been found to respond to busulfan treatment with a decreasing frequency of Ph¹ positive metaphases in the bone marrow. Such cases have, in general, been hypoplastic at the time of investigation.¹⁷,¹⁸

In relapse, the trisomy 8 cell population of our patient again dominated the bone marrow, but now with the additional double translocation [t(2;18), t(9;15)]. It seems likely that the mechanism behind this change in the karyotype of the leukemic cells is the same as that of the sequential nonrandom evolution of abnormal malignant karyotypes previously demonstrated in rat sarcomas.¹⁹,²⁰

Further studies have to clarify whether trisomy C in acute myeloblastic leukemia and sideroblastic leukemia is always due to an extra number 8 chromosome and whether such cases have specific features as to response to treatment and prognosis.

REFERENCES

Trisomy 8 in Acute Myeloblastic Leukemia and Sideroachrestic Anemia