Chronic Myelodysplastic Syndrome (Preleukemia) With the Philadelphia Chromosome

By Daniel G. Roth, Carol M. Richman, and Janet D. Rowley

A patient with severe anemia, reticulocytopenia, and erythroid hyperplasia of the bone marrow developed fatal acute nonlymphocytic leukemia after 3 yr. A Philadelphia chromosome with the typical 9/22 translocation t(9q+;22q-) was identified by banding techniques in a small number of bone marrow cells throughout the preleukemic phase of the illness (14%-38% of metaphases) and during the acute transformation (50%). Granulocytic colony formation in vitro was abnormal in the preleukemic phase. The diagnosis of chronic granulocytic leukemia was excluded on the basis of clinical and laboratory findings. The identification of the Ph' chromosome in this form of chronic myelodysplastic syndrome (preleukemia) provides a new example of a hematologic disorder predisposing to acute leukemia in which this chromosomal abnormality occurs.

The Philadelphia (Ph') chromosome is present in the hematopoietic cells of most patients with chronic myelogenous leukemia (CML) and has been reported only rarely in other disorders. In most instances, the cytogenetic abnormality involves a translocation of material from the long arm of chromosome 22 to the long arm of chromosome 9, present in 100% of bone marrow cells. Variant forms of the Ph' chromosome have been identified in patients with CML in whom translocation to a chromosome other than no. 9 occurs.

We report on a patient with Ph'-positive chronic myelodysplastic syndrome (preleukemia) in whom the clinical findings of CML were absent. A typical Ph' chromosome was present in a subpopulation of bone marrow cells for several years prior to the development of acute leukemia.

**MATERIALS AND METHODS**

The bone marrow chromosomes were prepared and analyzed as described previously. Metaphase cells were stained with Leishman-Giemsa and were photographed; the same cells were destained, then stained with quinacrine mustard and rephotographed with ultraviolet fluorescence. Chromosomes were identified according to the Paris nomenclature.

Bone marrow granulocytic progenitor cells (CFU-C) were assayed in methylcellulose as previously described. Ficoll-Hypaque-separated marrow mononuclear cells were plated in triplicate at a final concentration of 10³ cells/ml. Colony-stimulating activity (CSA) was provided by leukocyte-conditioned medium prepared with 1% phytohemagglutinin (PHA-LCM). After 14 days of incubation, colonies (>20 cells) were counted, and the results were expressed as mean CFU-C ± standard error of the mean.

**CASE REPORT**

The 49-yr-old white female patient was referred to the University of Chicago Hospitals in September 1976 for evaluation of macrocytic anemia of several months' duration that was unresponsive to B₁₂ and folate therapy. The findings in the physical examination were unremarkable except for pallor. There was no adenopathy or hepatosplenomegaly. Blood counts included a hemoglobin of 8.2 g/dl, hematocrit 26%, reticulocytes 1.1%, platelets 425,000/μl, and a white cell count 6500/μl, with 53% neutrophils, 35% lymphocytes, 9% monocytes, 2% eosinophils, and 1% basophils. Red cell indices were normal except for a mean corpuscular volume of 112 fl. Additional laboratory studies, with normal results, included electrolytes, liver and renal function tests, thyroid functions, prothrombin time, and partial thromboplastin times, serum complement, and antinuclear antibodies. The serum cholesterol was slightly reduced at 160 mg/dl. The serum folate level was 15.4 ng/ml, vitamin B₁₂,1100 pg/ml, iron 205 μg/dl, and iron binding capacity 330 μg/dl. The leukocyte alkaline phosphatase score was 56 (normal 43-123). A Schilling test without intrinsic factor gave normal results, with 10.6% excretion in 24 hr.

Examination of the bone marrow at this time showed 80% cellularity, a myeloid:erythroid ratio of 2:1, and mild megaloblastic changes in early erythroid precursors. The karyotype analysis of this and subsequent marrow samples is summarized in Table I. Five of eight cells in metaphase had a normal karyotype, and three contained a typical Philadelphia chromosome (Fig. 1). There was no response to therapeutic trials of pyridoxine, androgens, or prednisone. A diagnosis of chronic myelodysplastic state ("preleukemia") was made, and the patient was treated symptomatically with red cell transfusions approximately every 4-12 wk.

One year later, in August 1977, the symptoms and the results of a physical examination were unchanged. The white blood cell count was 7200/μl with a normal differential, and the platelet count was 366,000/μl. A bone marrow sample was 90% cellular with a myeloid:erythroid ratio of 5:1. The karyotype showed 31% Ph'-positive cells.

Two years after diagnosis, in August 1978, the white blood cell count was 15,200/μl, but the differential count remained normal and the platelet count was 406,000/μl. The patient had been maintained on prednisone, 20 mg/day. The bone marrow showed hypercellularity and an increased myeloid:erythroid ratio of 10:1. The karyotype of this sample showed 14% cells with a Ph' chromosome; the remaining cells had a normal chromosome pattern. The bone marrow was assayed for CFU-C at this time. With PHA-LCM as a source of CSA, the patient's marrow had 6 ± 2 colonies.
Table 1. Summary of Cytogenetic Analyses of Bone Marrow Cells

<table>
<thead>
<tr>
<th>Date</th>
<th>Chromosome Number</th>
<th>Total Cells</th>
<th>Number of Ph' Cells (%)</th>
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<tbody>
<tr>
<td>9-30-76</td>
<td>&lt;45' 45' 46 47</td>
<td>8 (8)</td>
<td>3 (38)</td>
</tr>
<tr>
<td>8-19-77</td>
<td>5 2 14 25</td>
<td>23 (13)</td>
<td>4 (31)</td>
</tr>
<tr>
<td>8-16-78</td>
<td>3 7 17 7</td>
<td>27 (14)</td>
<td>2 (14)</td>
</tr>
<tr>
<td>2-14-79</td>
<td>2 2 7</td>
<td>11 (8)</td>
<td>3 (38)</td>
</tr>
<tr>
<td>5-9-79</td>
<td>1 1</td>
<td>2 (2)</td>
<td>1 (50)</td>
</tr>
</tbody>
</table>

*Cells with less than 46 chromosomes were lacking various chromosomes due to cell breakage.
†The numbers in parentheses indicate the number of cells analyzed with banding.
‡Calculated as percent of cells analyzed with banding.
§One cell + 8, one ambiguous.

compared to 187 ± 2 for a normal control marrow. The range of normal bone marrow CFU-C in our laboratory is 50–200/10^3 cells. In October 1978, after the prednisone had been tapered off and discontinued, the white cell count was 6000. In December 1978, it was 5500. The leukocyte alkaline phosphatase at this time was 170.

In May 1979, the patient was hospitalized with severe anterior chest and bilateral hip pain. During the preceding 2 mo, she had become progressively weaker despite transfusions. Marked tenderness to palpation was present over the right second and third ribs. There was no hepatosplenomegaly. The hematocrit was 27%, platelets 19,000, and white cell count 2800, with 29% neutrophils, 6% bands, 51% lymphocytes, 7% monocytes, 3% metamyelocytes, and 4% blasts. Blood chemistry findings were normal except for an alkaline phosphatase of 147 IU/ml and a leukocyte alkaline phosphatase of 160. A bone marrow aspirate was unsuccessful, but a bone core biopsy showed a hypercellular marrow with increased reticulin and virtually total replacement of normal elements with myeloblasts. Cytogenetic studies of material from the core biopsy sample again showed the Ph' chromosome, with no additional abnormalities.

Chemotherapy with cytosine arabinoside, 6-thioguanine, and daunomycin was followed by worsening of the pancytopenia. The white cell count fell to zero and the platelets to 2000. The patient developed fever, and infiltrates were noted in the lower and upper lobes of the right lung. Antibiotics were administered together with granulocyte and platelet transfusions. A bone marrow biopsy specimen obtained 10 days after completion of the chemotherapy was 20% cellular and showed persistence of leukemia. Despite continued supportive care, the patient died on June 8, 1979. Permission for an autopsy was refused.

**DISCUSSION**

The clinical and laboratory observations on this patient were consistent with a chronic myelodysplastic syndrome, so-called “preleukemia.” However, the presence of the Philadelphia chromosome with the usual 9/22 translocation in this patient required consideration of a diagnosis of CML, even though the clinical and laboratory findings were entirely inconsistent with CML. At the time of diagnosis and throughout the course of the disease, the major finding was a severe refractory anemia with ineffective erythropoiesis. Splenomegaly was absent, and increased basophils were never observed. The leukocyte alkaline phosphatase was normal on three occasions. The Ph' chromosome was present only in a small percentage of cells, whereas it is usually found in 100% of the metaphases in CML. Finally, the CFU-C results are not typical of CML. The concentration of CFU-C in CML marrows generally ranges from normal to very high values. In Goldman's study, 7 of 8 CML patients had colony counts 1–5 times higher than their upper limit of normal. In contrast, colony formation in preleukemia resembles that observed in acute myelogenous leukemia, that is, no or very few colonies are found. A maximum of 12 CFU-C/10^5 cells were observed by Senn in his serial studies of three preleukemia patients. (Normal marrow CFU range in his laboratory was 80–120/10^5 cells.) In the present case, the low concentration of CFU-C (6 ± 2) is most consistent with a diagnosis of preleukemia.
The Philadelphia chromosome has been reported in a variety of conditions other than chronic myelogenous leukemia, including acute leukemia without an antecedent chronic phase, and other myeloproliferative syndromes. In many such instances, chromosome banding studies were not done, and the certainty of identification of the Ph' is in some doubt. In other cases, CML with somewhat atypical features may have been present. It is well established, however, that the Ph' chromosome may occur in patients with acute leukemia who have no history of a preceding chronic phase.

The Ph' chromosome has been reported by Canellos and Whang-Peng in a patient with a preleukemia state different from that described here. Their patient had leukocytosis and granulocyte hyperplasia in the bone marrow, with no other findings of CML. The Ph' chromosome was initially present in a small number of cells. After 5 yr, acute leukemia developed, with 100% Ph'-positive cells.

The finding of the Philadelphia chromosome, documented in serial samples by banding techniques, in the more common form of preleukemia with refractory anemia and hypercellular marrow expands the association between this cytogenetic abnormality and disorders other than CML. Although it was sometimes difficult to obtain an adequate number of mitotic cells for analysis, the patient’s marrow always contained a mixture of normal and Ph'-positive cells. No new chromosome changes were observed during evolution of the patient’s disease. The relationship between the presence of the Ph' chromosome and the eventual occurrence of acute leukemia remains an important unresolved problem.

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REFERENCES

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