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New reciprocal translocation t(5;10)(q33;q22) associated with atypical chronic myeloid leukemia

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ABSTRACT

We report a new chromosomal reciprocal translocation t(5;10)(q33;q22) in a 49-year-old man with atypical chronic myeloid leukemia (a-CML) and history of occupational exposure to petroleum products including benzene and other hydrocarbons. The t(5;10)(q33;q22) was found in 94% and 84% of metaphases in peripheral blood and bone marrow cells, respectively. Cytogenetic analysis of single colonies derived from granulocyte-macrophage (CFU-GM), and erythroid (BFU-E) hematopoietic progenitors showed that 88% and 40% of CFU-GM and BFU-E, respectively, had the t(5;10)(q33;q22). In contrast, peripheral blood T-lymphocytes, and cutaneous fibroblasts had normal 46,XY karyotype. Molecular analysis of the t(5;10)(q33;q22) translocation breakpoint is currently underway in order to identify genes located in this region which might provide insights into the pathogenesis of atypical myeloproliferative disorders.

Key words: chromosomal translocation t(5;10)(q33;q22), atypical chronic myeloid leukemia

Chronic myeloproliferative disorders (MPD) are clonal diseases of the hematopoietic stem cell due to acquired somatic mutations resulting in a selective growth advantage of the neoplastic clone over normal elements, and characterized by progression to acute leukemia. Despite distinct pathologic entities with well defined clinical features among chronic MPDs, these diseases are characterized by a wide biological heterogeneity, suggesting that increased proliferation of hematopoietic cells is supported by a variety of still unidentified molecular mechanisms.

Clonality analyses at the level of hematopoietic progenitors have indicated heterogeneity of stem cell involvement in acute MPD but not in chronic MPD. Cytogenetic and molecular studies of recurrent chromosomal translocations occurring in chronic MPD are contributing to uncovering pathogenetic mechanisms of leukemogenesis as well as identifying new treatment strategies.[1] We report here a new chromosomal translocation, occurring at the level of an early hematopoietic progenitor, associated with distinctive clinical and cytogenetic features supporting the diagnosis of atypical chronic myeloid leukemia (a-CML).

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Case Report

Clinical features

A 49-year-old man (UPN 54375) was referred to the Department of Oncology-Hematology, Istituto Clinico Humanitas, Milan, Italy, because of generalized bone pain and leukocytosis. *Family history*: negative for malignant diseases. *Personal history*: occupational exposure to petroleum products including benzene and other hydrocarbons for 27 years. The recent complaint leading to hospital admission was generalized bone pain increasing from mild to severe during the preceding 4 weeks. *Physical examination*: grade 3 (WHO/ECOG Scale) deterioration of performance status because of bone pain. Spleen, liver, and lymph nodes were within normal limits.

Hematologic findings

Complete blood counts: leukocytes 33×10^9/L, erythrocytes 3.58×10^9/L, reticulocytes 57,130/µL, Hb 10.3 g/dL, platelets 398×10^9/L. *Peripheral blood smear* (MGG staining): nucleated cells were 6% erythroblasts, 8% lymphocytes, 3% monocytes, 6% eosinophils, 2% basophils, 30% neutrophils, 10% metamyelocytes, 6% myelocytes, 26% promyelocytes, and 3% blasts. Myeloid cells had remarkable granulocytic dysplasia with increased size, reduced N/C ratio, and hybrid eosinophilic and basophilic granulations of increased size and number. There was anisocytosis (RDW 20%) and poikylocytosis (schistocytes and dacrocytes) of erythrocytes; platelets were normal with occasional giant elements. Leukocyte alkaline phosphatase score was reduced to 10 (n. v. = 15-100). *Bone marrow smear* (MGG staining): the first

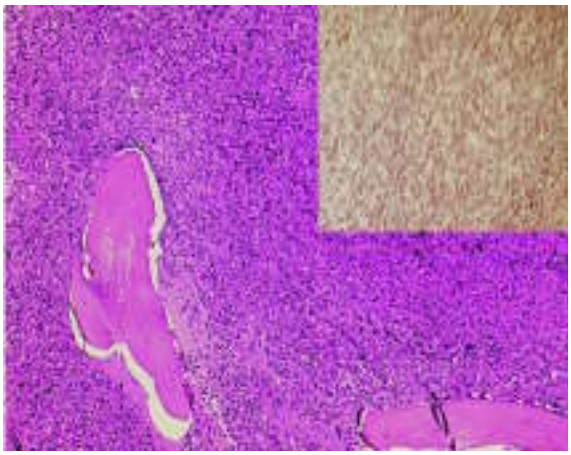

Figure 1. Bone marrow biopsy from posterior iliac crest demonstrating the characteristics of a myeloproliferative disorder with remarkable myeloid hyperplasia and loss of fat space (hematoxylin-eosin staining, ×4 magnification), and focally increased reticulin framework (Gomori staining, inlet photograph, ×2 magnification).

Table 1. Phenotype of bone marrow and peripheral blood cells at time of presentation and accelaration of a-CML.

Lineage specificity of monoclonal antibody	*Percentage of positive cells*		
		Peripheral blood	
	Bone marrow	*Presentation*	*Acceleration*
Hematopoietic progenitor cells			
CD34	0.9	0.5	1.4
Myeloid cell lineage			
CD11b	9.5	61	57
CD13	10	67	51
CD15	10	65	ND
CD33	13	66	43
T-cell lineage			
CD2	21	16	26
CD3	20	13	21
CD4	13	8	16
CD7	24	17	25
CD8	9	5	10
CD25	0.8	1	1
B-cell lineage			
CD10	0.5	ND	0.1
CD19	3.5	3	4
CD22	4	ND	3
CD23	7	4	8
Natural killer cells			
CD56	19	8	ND
CD3-CD16-CD56+	5	5	3
Miscellaneous			
HLA-DR	9	13	9

Percentage of positive cells was assayed by direct immunofluorescence with commercially available monoclonal antibodies (Becton-Dickinson, San José, CA, USA) using a FACSort flow cytometry system. ND, not determined.

bone marrow aspirate was *punctio sicca*. The second bone marrow aspirate, upon reduction of leukocytosis with hydroxyurea treatment, was without bone spicules and showed myeloid hyperplasia (M/E ratio 15/1) comprising myeloid cells with granulocytic dysplasia analogous to cells found in peripheral blood. There were 25% and 15% promyelocytes and eosinophilic precursor-mature cells, respectively. Erythroid cells had cytoplasm and nuclei at dissimilar stages of maturation and occasional intercellular bridges. Megakaryocytes were normal. *Bone marrow biopsy:* HE, MGG, and reticulin staining showed marked hypercellularity with loss of fat spaces, M/E ratio 20/1, myeloid precursors with dysplastic features at different stages of maturation although mainly immature, mild increase of megakaryocytes, and focally increasead reticulin framework (Figure 1). *Immune phenotype of bone marrow and peripheral blood:* bone marrow and peripheral blood at the time of presentation as well as at acceleration comprised heterogenous cell populations, thus excluding the progression to acute leukemia (Table 1). *Evaluation of hematopoietic progenitors:* evaluation of hematopoietic progenitors circulating in peripheral blood revealed increased growth of multilineage CFU-Mix (100±40/mL), erythroid BFU-E (3,425±283/mL), and granulocyte-macrophage CFU-GM (5,725±140/mL) progenitors (n. v. = CFU-Mix 60±10/mL, BFU-E 430± 50/mL, CFU-GM 520± 70/mL) and was consistent with a growth pattern typical of MPD.

Cytogenetic findings

The majority of metaphases of mononuclear bone marrow and peripheral blood cells exhibited the reciprocal translocation t(5;10)(q33;q22) (Figure 2). As summarized in Table 2, karyotype analysis of single colonies relative to CFU-GM and BFU-E showed that the t(5;10)(q33;q22) was limited to 88% and 40% of these lineages, respectively. In contrast, T-lymphocytes and cutaneous fibroblasts had normal 46, XY karyotypes (Table 2).

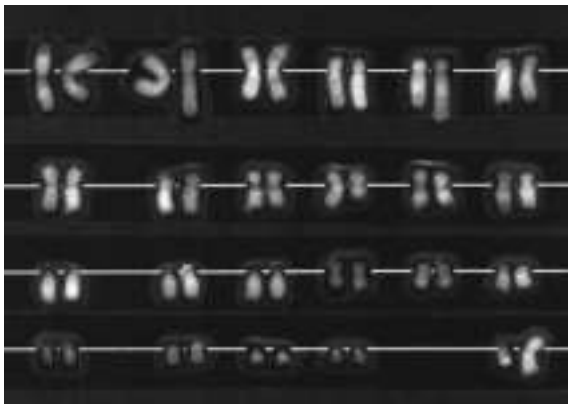

Figure 2. Representative karyotype of a Q-banded metaphase from bone marrow aspirate at presentation of disease. Arrows point to derivative chromosomes resulting from t(5;10)(q33;q22).

Table 2. Summary of cytogenetic findings.

Tissue and type of cells	*Karyotype*	*Abnormal metaphases*
Bone marrow		
Mononuclear cells	46, XY [8]	
	46, XY, t(5;10)(q33;q22) [41]	84%
Peripheral blood		
Mononuclear cells	46, XY [1]	
	46, XY, t(5;10)(q33;q22) [17]	94%
CFU-GM progenitors*	46, XY [3]	
	46, XY, t(5;10)(q33;q22) [22]	88%
BFU-E progenitors*	46, XY [15]	
	46, XY, t(5;10)(q33;q22) [10]	40%
T-lymphocytes° (Immune magnetic sorted and PHA-stimulated MNCs)	46, XY [5]	0%
Skin		
Fibroblasts	46, XY [16]	0%

Figures in [ ] represent the number of metaphases analyzed by QFQ binding, with standard methods.
**Single colony karyotyping relative to CFU-GM and BFU-E hematopoietic progenitors was carried out as previously described.[11] °T-lymphocytes were sorted by MiniMACS (Mylteni Biotec, Germany).[12] ND, not determined.*

Molecular findings

Reverse transcriptase polymerase chain reaction (RT-PCR)[11] for $p210^{BCR\text{-}ABL}$ and $p190^{BCR\text{-}ABL}$ did not show the BCR-ABL rearrangement.

Diagnosis and follow-up

The above clinical and laboratory findings fitted the FAB criteria of a-CML[2] presenting in accelerated phase.[3] Administration of hydroxyurea 1-2 g qd po resulted in induction of a chronic phase as shown by decreased WBC counts, decrease of generalized bone pain from severe to mild, and improvement of performance status from grade 3 to grade 1. After 18 weeks of treatment with hydroxyurea, patient progressed to a second accelerated phase with increase of WBC to 24×10^9/L, severe generalized bone pain, and grade 4 deterioration of performance status. Administration of cytosine arabinoside 2,000 mg iv and prednisone 75 mg po qd for 7 days resulted in induction of a second chronic phase that is presently maintained with combined hydroxyurea and 6-thioguanine therapy.

Discussion

We report here a new t(5;10)(q33;q22) detected in hematopoietic progenitor and precursor cells but not in T-lymphocytes and cutaneous fibroblasts of an individual with a-CML. The data presented are consistent with the hypothesis of a neoplastic event occurring in an early hematopoietic progenitor cell and are in keeping with clonality data obtained in classical MPD. Other reported translocations involving the long arms of the same chromosomes, i.e., a t(5;10) (q35;q23 or 24) in a child with M4 ANLL[4] and a t(5;10)(q13;26) in a man with M5a ANLL,[5] are not related to the present abnormality because of different clinical, hematologic, and cytogenetic characteristics.

The t(5;10)(q33;q22) presented here involves chromosomal regions containing genes implicated in the pathogenesis of various malignancies.[1] The chromosome 10q22 contains tumor suppressor genes such as the juvenile polyposis syndrome locus JP1 and the PTEN/MMAC1/TEP1 gene responsible for the familial cancer syndromes Cowden disease and Bannayan-Zonana syndrome and genes that may also be somatically mutated in several types of sporadic cancers such as glioblastoma multiforme, thyroid, endometrial, and prostate carcinomas.[6] The chromosome 5q33 contains the platelet-derived growth factor β receptor (PDGFβR) gene which encodes a member of the tyrosine kinase receptor family. The translocations t(5;12) (q33;p13), t(5;7)(q33;q11.2), and t(5;14) (q33;q32), occurring in subsets patients with chronic myelomonocytic leukemia (CMML)[7,8] and acute non-lymphocytic leukemia,[9] cause the fusion of PDGFβR gene with either TEL gene on chromosome 12p13, HIP1 gene on chromosome 7q11.2, or CEV14 gene on chromosome 14q32. The TEL/PDGFβR,[7] HIP1/PDGFβR,[8] and CEV14/PDGFβR[9] fusion genes result in the constitutive oligomerization and activation of PDGFβR tyrosine kinase activity leading to transformation of hematopoietic progenitor cells. Patients with CMML and ANLL and the above translocations share remarkable eosinophilia with the present patient with t(5;10) (q33;q22). Eosinophilia also occurs in other hematopoietic malignancies affecting chromosome 5q.[9]

The diagnosis of a-CML comprises a small group among MPDs which are heterogeneous with respect to morphologic and clinical features. A common denominator is the absence of Philadelphia chromosome, as documented in the present case. Although the clinical phenotype of a-CML of the patient with t(5;10)(q33;q22) in this report differs from that of other patients with CMML and translocations involving the 5q33, one should consider that debate exists as to whether a-CML and CMML are separate disorders or part of a spectrum of myeloproliferative disorders with dysplastic features.[10]

In our patient, based on the presence of suppressor genes on chromosome 10q22, it may be speculated that the a-CML occurred in the context of a multistep carcinogenesis whose hypothetical triggering may have been exposure to petroleum products. Molecular cloning of the breakpoints involved in the t(5;10) (q33;q22) translocation reported in this article is presently underway and is expected to identify the

genes located in this region which may provide further insight into the pathogenesis of atypical myeloproliferative disorders.

Contributions and Acknowledgments

SS: diagnosis and therapy, coordination of research studies and writing the article; GS: cytogenetics; RS: flow cytometry; AN: clinical care and morphology; MR: morphology; CM: cytogenetics and molecular biology assays; AS: clinical care; CC-S: diagnosis and therapy, coordination of research studies, and writing the article.

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Disclosures

Conflict of interest: none.

Redundant publications: no substantial overlapping with previous papers.

Manuscript processing

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