Translocation t(12;13)(p13;q14) in a patient with imatinib-sensitive MDS/MPD associated with resistance to treatment: review of the literature

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The category of myelodysplastic syndromes/myeloproliferative diseases (MDS/MPD) is a relatively new group of malignant hematologic diseases developed by the World Health Organization. These hematologic disorders lack the BCR/ABL fusion gene, although they can be associated with chromosomal translocations that involve genes encoding other protein kinases. Imatinib mesylate was recognized as a potent inhibitor of some of those kinases. We present a patient with a previously treated acute myeloid leukemia, who, after a 9-year-long remission, developed an MDS/MPD with normal karyotype, which initially responded to imatinib mesylate. Translocation t(12;13)(p12;q14) was detected after loss of response to imatinib treatment. Translocation t(12;13) is rare. It has been described in several hematologic malignancies including chronic myelomonocytic leukemia but not in MDS/MPD, previously described as Philadelphia-negative chronic myelogenous leukemia. Moreover, the correlation of this molecular abnormality with loss of efficacy of imatinib is unique in the literature. ***Anti-Cancer Drugs*** **22:944–947** © **2011 Wolters Kluwer Health | Lippincott Williams & Wilkins.**

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Introduction

Imatinib mesylate, first described as a BCR/ABL tyrosine kinase inhibitor, is a potent inhibitor of several other kinases, molecular abnormalities of which are found in a vast range of hematologic disorders [1,2]. Myelodysplastic syndromes/myeloproliferative diseases (MDS/MPD) comprise a rather new hybrid category of hematologic disorders according to the World Health Organization classification [3], with clinical and laboratory characteristics of both MDS and MPD. Response to imatinib has been reported in patients with MDS/MPD bearing several translocations that involve genes encoding for kinases other than BCR/ABL [1,2]. The case presented below is that of a patient, previously treated for acute myeloid leukemia (AML), who developed an MDS/MPD showing a favorable response to imatinib mesylate without any of the molecular abnormalities that are known to be responsive to the drug and who became resistant to the drug after developing the translocation t(12;13).

Case presentation

A 58-year-old lady presented to our Outpatient Hematologic Unit in November 1994 because of 2-month fatigue and malaise as well as 1-week spontaneous bruising and gingival hemorrhage. Her medical history included colon polyps, irritable bowel syndrome, and osteoporosis, whereas family and social histories were unremarkable. Physical examination revealed paleness and petechiae in the oral cavity and ecchymoses at her extremities. No lymph node enlargement or organomegaly was noted. The results of the complete blood count were: hemoglobin (Hb) 9.7 g/dl, white blood cells (WBC) 40×10^9/l with 23% blasts, and platelets (PLT) 55×10^9/l. Bone marrow aspiration showed hypercellularity with 50% blasts, positive for CD13, CD14, CD33, CD34, human leukocyte antigen D-related (HLA-DR), and CD2. Cytogenetics showed no abnormal findings. FAB (French–American–British) AML-M1 was documented and the patient was treated according to the protocol of the Greek Hematology Association with mitoxantrone, aracytine, and etoposide (induction), and with aracytine (consolidation). In June 1995, bone marrow examination showed that she was in complete hematologic remission, which lasted for the next 9 years.

In November 2004, a borderline increase (5–6%) of the bone marrow blasts suggested a possible relapse. Over the

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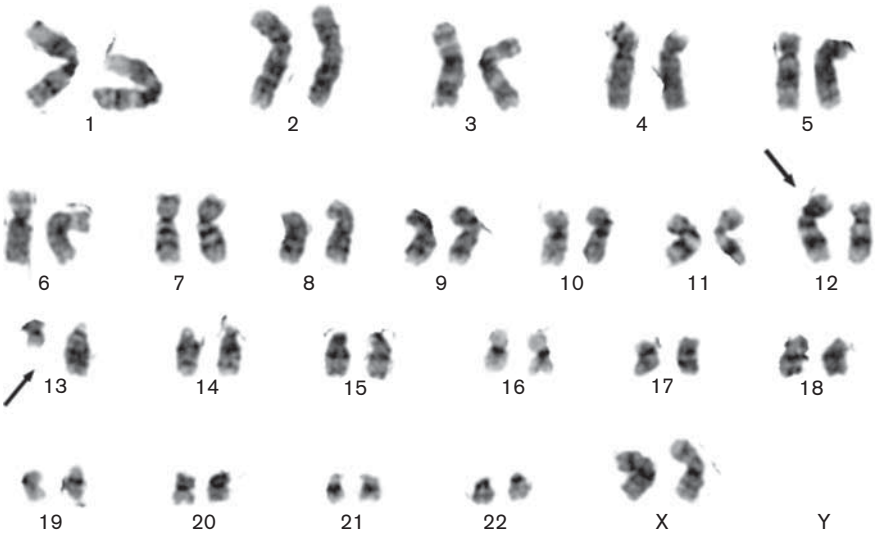
following year, serial blood counts showed an increasing WBC count (up to $15\,600 \times 10^9$/l), anemia (hematocrit = 32%), and thrombocytopenia (PLT = 131×10^9/l), whereas bone marrow examination showed a highly hypercellular bone marrow with complete predominance of granulocytes with 5–6% bone marrow blasts and elimination of erythroid precursors and megakaryocytes. Cytogenetic analysis was normal. PCR studies for the three transcripts (p190, p210, and p230) of BCR/ABL were negative. The patient was also PCR negative for ETV6/ABL and for platelet-derived growth factor receptor α (PDGFRα) and PDGFRβ fusion genes. A diagnosis of MDS/MPD was made and the patient was treated with hydroxyurea and received packed red blood cell transfusions every 2 weeks. Six months later, she presented with Coombs-positive hemolytic anemia (hematocrit = 19%). All specific laboratory tests for known causes of autoimmune hemolytic anemia were negative. The patient was treated with 48 mg of methylprednisolone daily orally. Because of further deterioration of the hematocrit, she was given 500 mg of cyclophosphamid intravenously, followed by 50 mg twice daily orally, and because of lack of response vincristine and danazole were added. After autoimmune hemolytic anemia was resolved, her WBC count started increasing (100×10^9/l) despite hydroxyurea administration, with concomitant worsening of thrombocytopenia. A new cytogenetic study was carried out in January 2007, which revealed a normal karyotype in 25 examined mitoses. In February 2007, the patient was given 400 mg of imatinib mesylate daily orally, with eventual normalization of the WBC and PLT counts. Imatinib was discontinued 2 months later because of grade IV thrombocytopenia and leucopenia, followed by hematologic normalization for 6 months. A routine bone marrow aspiration showed normal cellularity and blast cells within normal range. No cytogenetic analysis was carried out at this time point because of a normal karyotype before starting treatment with imatinib.

A gradual decrease in the hematocrit and PLT count was noted during the following 5 months, and a concomitant leukocyte increase. Reevaluation of the patient showed no signs of autoimmune hemolytic anemia or autoimmune thrombocytopenia. The patient did not show a favorable response to a second treatment with imatinib mesylate. At this period, the level of imatinib mesylate was measured in the international reference laboratory of Novartis Company and found to be within therapeutic range. Bone marrow examination confirmed the diagnosis of MDS/MPD. Repeated cytogenetic analysis revealed 46,XX t(12;13)(p13;q14) abnormality (Fig. 1) in 75% of examined mitoses. Interphase fluorescence *in situ* hybridization analysis was carried out using a commercially available DNA probe for 13q14 (D13S319 Spectrum-Orange Probe; Vysis, Downers Grove, Illinois, USA). It revealed deletion of the 13q14 band in 75% of nuclei (Fig. 2). Further molecular studies were carried out, including detection of rearrangements of the FLT3 gene, ETV6/ABL, and PDGFRα and PDGFRβ fusion genes, and were all found to be negative. Moreover, fluorescence *in situ* hybridization analysis was carried out to examine the genetic locus of the retinoblastoma (RB1) gene on

Fig. 1

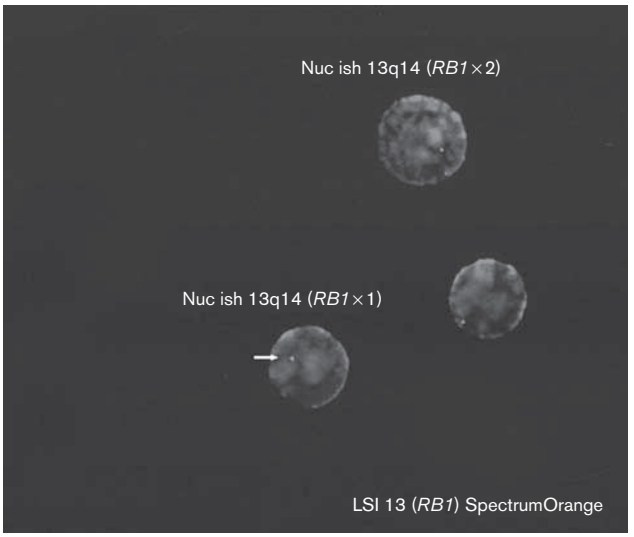


Cytogenetic study. The analysis was conducted by standard G-banding; karyotypes were interpreted using the International System for Human Cytogenetic Nomenclature criteria (2005). A minimum of 25 mitotic cells were examined. The following karyotype was observed: 46,XX,der(12)t(12;13)(p13;q14),der(13)t(12;13)(p13q14) in 75% of the mitoses examined.

Fig. 2

Interphase fluorescence *in situ* hybridization analysis, using a DNA probe for 13q14 (D13S319 SpectrumOrange Probe), revealed deletion of the 13q14 band in 75% of the nuclei.

Fig. 3



Interphase fluorescence *in situ* hybridization analysis, using the LSI 13 [retinoblastoma (*RB1*)] 13q14 SpectrumOrange probe) revealed loss of the *RB1* gene in 80% of the examined nuclei. Two hundred nuclei were examined. The two red fluorescent signals indicate normal nuclei (40 of the 200 nuclei, 20%), whereas in 160 nuclei (80%), there is only one fluorescent signal (arrow), indicating loss of the genetic locus of the *RB1* gene on chromosome 13.

chromosome 13, using the LSI 13(*RB1*) 13q14 SpectrumOrange probe. The test was positive for del(13)(q14)(*RB1*-) in 80% of the examined nuclei (Fig. 3).

On her last admission, she suffered an upper gastrointestinal hemorrhage and a severe pulmonary infection and died in February 2009. The patient was not treated with a second-generation tyrosine kinase inhibitor because of lack of a relevant clinical trial.

Discussion

MDS/MPD is a relatively new hybrid category of malignant hematologic diseases developed by the World Health Organization. These hematologic disorders lack the BCR/ABL fusion gene, although they can be associated with chromosomal translocations that involve genes encoding protein kinases, including the ABL-related protein, auxin-regulated gene (ARG), the KIT receptor tyrosine kinase, PLT-derived growth factors α and β (PDGFRα, PDGFRβ), and c-Fms. Imatinib mesylate was recognized as potent inhibitor of those kinases [1].

In the literature, several novel fusion proteins have been implicated in the pathogenesis of chronic myeloproliferative disorders such as ETV6-ABL, PDGFRα, and PDGFRβ hybrids.

ETV6-ABL: ETV6 is an E-twenty six-related transcription factor, encoded on chromosome 12p12-13, and the only non-BCR fusion partner identified for ABL; it was first described in a child with acute lymphoblastic leukemia (ALL), and it was subsequently reported in several patients with BCR-ABL-negative atypical chronic myelogenous leukemia (CML). One of them was a 36-year-old man with findings consistent with CML in blast crisis. He was treated with induction therapy for AML with minor cytogenetic response. Treatment with imatinib was started and the patient remained stable for 3 months. Cytogenetic analysis revealed an additional t(12;13)(p12;q13), as in our patient, with the 12p breakpoint proximal to ETV6. The patient relapsed into blast crisis and died shortly afterward [2,4]. In one case of a myeloproliferative disorder with hypereosinophilia and a t(12;13)(p13;q12) translocation, ETV6 was found to be fused with the FLT3 gene [5]. Other cases of translocations involving 12p13 but not ETV6 rearrangement have already been reported in Philadelphia-positive CML patients, who acquired it as a second abnormality just before blast transformation.

Novel PDGFRβ fusion genes: TEL/ETV6-PDGFRβ is by far the most frequent with more than 30 cases described [6], whereas other fusions include HIP1–PDGFRβ, RAB5–PDGFRβ, KIAA1509–PDGFRβ, H4–PDGFRβ, PDE4DIP–PDGFRβ, NIN–PDGFRβ, and TP53BP1–PDGFRβ.

Novel PDGFRα fusion genes: In patients with hypereosinophilic syndrome and atypical chronic myeloid leukemia, response to imatinib has been attributed to chromosomal aberrations involving PDGFRα [7].

In our case, a patient with a previously treated AML, after remission for 9 years, developed MDS/MPD with a normal karyotype and without ETV6-ABL rearrangement, or rearrangement of PDGFRα, and PDGFRβ, which initially responded to imatinib mesylate. Translocation t(12;13)(p12;q14) was detected after loss of response to imatinib treatment. As the presence of t(12;13) was documented during the final reevolution of the disease, with acquired resistance to imatinib, we assume that it may

not be responsible for the MDS/MPD pathophysiology in our case but for resistance in the specific treatment. Translocations between chromosomes 12 and 13 are generally rare. They have been described in ALL, AML, CML, chronic lymphocytic leukemia, and MDS, including a chronic myelomonocytic leukemia, but not in MDS/MPD, which was previously reported as Philadelphia-negative CML. At least 35 cases of t(12;13) have been reported already, with breakpoints cytogenetically scattered from 12p11 to 12p13 and from 13q11 to 13q34 [8–10].

On searching the literature separately for the two chromosomes and for the locations of the breakpoints (12p13 and 13q14) participating in the translocation of our patient, we found the following. Chromosomal abnormalities of the short arm of chromosome 12 have been observed in ALL, acute nonlymphoblastic leukemia, and MDS. The breakpoints in 12p are very heterogeneous and the delineation of subcategories of MDS or acute nonlymphoblastic leukemia is rather difficult [11]. One of the most common breakpoints in 12p is 12p12-13, where ETV6 is located.

Rearrangements of 13q12-14 could be related to the initiation of the disease (sole abnormality in some cases) and to its progression (additional abnormality in other cases) [12]. Molecular analysis has shown that monoallelic loss of the RB1 gene, located on 13q14, occurs in up to 34% of chronic lymphocytic leukemia cases [13,14], but commonly the 13q14 deletions do not lead to an inactivation of this gene. Loss of the RB1 gene has been commonly described in chronic lymphoproliferative diseases, less frequently in other hematologic clonal diseases such as AML, CML, MDS [15], and chronic myelomonocytic leukemia [16], but never in MDS/MPD. In cases of CML, inactivation of the RB1 gene has been described during blastic crisis [17], but this correlation has not been described in other chronic myeloproliferative disorders [18].

Conclusion

Our case is the first report in the literature showing that an MDS/MPD with t(12;13)(p13;q14) rearrangement develops as a second malignancy after AML, which remained in complete remission for 9 years. We speculate that etoposide was not the cause of the second malignancy because of the 9-year interval after AML treatment. This is also the first report that states that loss of sensitivity to imatinib treatment of an MDS/MPD with normal cytogenetics is directly associated with predominant appearance of a clone carrying t(12;13)(p13;q14) abnormality, associated with monoallelic loss of the RB1 gene, without evidence of AML transformation during the course of the disease.

Our patient remained in complete remission for 6 months after imatinib treatment, but the drug had probably a positive selective effect on the clone bearing the t(12;13)(p13;q14) rearrangement, which was resistant to treatment. Treatment with imatinib mesylate, or with second-generation kinase inhibitors, in cases with no proven sensitivity to the drug can improve clinical outcome and provide information about the biological and clinical importance of certain chromosomal abnormalities such as t(12;13)(p13;q14), particularly among younger patients, with other options of treatment such as bone marrow transplantation, who develop this abnormality.

Acknowledgements

Conflicts of interest

There are no conflicts of interest.

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