An unusual three-way translocation t(21;8;1)(q22;q22;q32) in a case of acute myeloid leukemia (M2)

Leucémie aiguë myéloïde (M2) avec une translocation complexe rare t(21;8;1)(q22;q22;q32) : à propos d’un cas

Abstract. Variant forms of the classic translocation t(8;21) are uncommon and account approximately 3% of all t(8;21)(q22;q22) in acute myeloid leukemia (AML) patients. These forms involve chromosomes 8, 21, and other chromosomes. Here we report a Tunisian patient with a complex rearrangement t(21;8;1)(q22;q22;q32) revealed by conventional chromosomal study at diagnosis. Fluorescence in situ hybridization study revealed the presence of the AML1-ETO chimeric gene on the derivative chromosome 8. To the best of our knowledge, this is the second case of t(21;8;1) of AML-M2 reported in the literature with the involvement of the same breakpoint at 1q32. This illustrates that this complex translocation is rarely encountered in AML and reinforces the fact that this region may harbour a critical gene candidate that may play an important role in the pathogenesis of AML. More cases are needed to elucidate its clinical features and prognosis.

Key words: AML-M2, t(21;8;1), conventional karyotype, FISH, AML1/ETO

Résumé. Les formes variables de la translocation classique t(8;21) sont rares et trouvées dans approximativement 3 % de tous les cas de la leucémie aiguë myéloïde (LAM) avec une translocation t(8;21)(q22;q22). Ces formes impliquent les chromosomes 8, 21 et d’autres chromosomes. Nous rapportons le cas d’un patient tunisien avec une translocation complexe t(21;8;1)(q22;q22;q32) révélée par caryotype en bandes R au moment du diagnostic. L’hybridation in situ fluorescente a révélé la présence du gène chimérique AML1-ETO sur le dérivé du chromosome 8. À notre connaissance, c’est la deuxième fois que cette translocation t(21;8;1) est décrite dans la littérature avec l’implication de la même région 1q32 chez un patient ayant une LAM-M2. Ceci illustre que cette translocation complexe est rarement rencontrée dans les LAM et renforce le fait que cette région peut héberger un gène candidat pouvant jouer un rôle important dans la pathogénie de la LAM. Plus de cas similaires sont nécessaires pour élucider ses caractéristiques cliniques et pronostiques.

Mots clés : LAM-M2, t(21;8;1), caryotype, FISH, AML1/ETO

The translocation (8;21)(q22;q22) is typically found in acute AML and is closely associated with French-American-British (FAB) M2 morphology [1]. This translocation is one of the most common karyotypic abnormalities in this disease, accounting for about 8–12% of patients with de novo AML [2]. The translocation generates a fusion gene located on the derivative chromosome 8, composed of the ETO gene located at 8q22 and the AML1 gene (also known...
as *RUNX1*) located at 21q22 and traduced on a unique chimeric leukemogenic protein [3, 4]. Variants of the t(8;21) are uncommon and accounts for approximately 3% to 4% of AML associated with t(8;21). It may display 3- or 4-way translocations involving 8q22 and 21q22. Although further accumulation of such cases are needed, it seems that the prognostic or clinicopathologic implications of variant t(8;21) might not differ from those of classic t(8;21) [5, 6]. Here we report a Tunisian patient showing a complex translocation t(21;8;1)(q22;q22;q32) with the involvement of the 1q32 breakpoint.

**Methods**

**Patient**

This 33-year-old male was hospitalized because of fever, weakness and on examination the patient looked pale. Complete blood cell analysis demonstrated a white blood cell count of 14.5 G/L with 50% blasts, a platelet count of 40 G/L, and a hemoglobin concentration of 36 g/L. The blasts were large and cytochemical staining showed the blasts to be positive for α-naphthyl butyrate esterase. Morphologically, the AML-M2 subtype according to the French-American-British (FAB) classification was present. The patient had relevant toxic professional exposure through his work as a painter. He was negative for HSV, hepatitis B and C, and HIV. Clinically, the case showed good prognosis. In deed, after receiving a standard course of induction therapy (idarubicin and cytosine arabinoside), the patient responded to treatment with complete remission. He further received 3 cycles of consolidation chemotherapy with a regimen of idarubicin and high-dose cytosine arabinoside C. He is now in complete clinical remission awaiting allogeneic bone marrow transplantation from his HLA-matched sibling.

**Conventional cytogenetic analysis and FISH**

At least 20 metaphase chromosomes were banded by the conventional RHG-banding technique performed on bone marrow (BM) cells of the patient, referred to our laboratory from the division of hematology of the university hospital of Sfax (Tunisia) as described previously [7]. Chromosomal abnormalities were described per the International System for Human Cytogenetic Nomenclature (ISCN) [8]. A dual-color FISH assay using *ETO* and *AML1*-specific probes (Vysis, Downers Grove, IL) was performed on bone BM cells, as previously described [9]. The *ETO* probe was directly labeled with SpectrumOrange, and the *AML1* probe was directly labeled with SpectrumGreen.

**Results**

Chromosome study using R-banding technique revealed a karyotype of 46,XY,t(21;8;1)(q22;q22;q32) in 15 metaphases analyzed. As shown in *figure 1*, we noted that the distal parts of the long arms of chromosome 1 and 8 were deleted and were both translocated to the long arm of chromosome 21.

FISH analysis for the *AML1/ETO* probe showed one normal orange (*ETO*) and one normal green (*AML1*) signals

![Figure 1. Partial R-banding karyotypes of the t(21;8;1)(q22;q22;q32) translocation. Normal chromosomes are shown on the left side and the derivative chromosomes on the right side. Arrowheads indicate breakpoints.](image-url)
on normal chromosomes 8 and 21 and one orange green fusion signal corresponding to the co-localization of ETO and AML1 signals on the rearranged chromosome 8 as, thus confirming the presence of the AML1/ETO fusion gene in this translocation. FISH results revealed also the presence of one small orange signal (ETO) on der(1) as shown in figure 2.

Discussion

Since 1976 [10], the FAB system was used for classifying AML, it was discovered that many cases of AML are associated with recurring genetic abnormalities that affect cellular pathways of myeloid maturation and proliferation. However, morphologic-genetic correlations are not always perfect, and the genetic findings may predict the prognosis and biologic properties of the leukemia more consistently than does morphology. Thus, some investigators suggest that a more relevant classification of AML which is the WHO classification [11]. According to this classification, patients with the specific recurring cytogenetic abnormalities t(8;21)(q22;q22), inv(16)(p13q22) or t(16;16)(p13;q22), and t(15;17)(q22; q12) should be classified as having AML regardless of the blast percentage. AML with the t(8;21)(q22;q22) is recognized as a distinct type of AML in the WHO classification. It often occurs in young patients and is associated with a favorable outcome [12-15]. Variant translocations of t(8;21) involving chromosomes 1, 2, 4, 5, 6, 7, 8, 10, 12, 13,15, 17, 18, 19, or 20 have been reported, with a frequency of ~3-4% [2, 16]. The prognosis associated these variants is still unclear. Nevertheless, some studies suggest that there are no obvious differences between complex and typical t(8;21) in regard to remission rate and disease free survival [17], whereas others indicate that the additional chromosome involved in the translocation could change the biological characteristics of leukemic cells, rendering it resistant to the cytotoxic effects of chemotherapeutic drugs [18]. Here we report a case of AML-M2 with a rare t(21;8;1)(q22;q22;q32). The involvement of chromosome 1 in t(8;21) has been previously reported in 8 cases in the literature including one other Tunisian patient with t(1;21;8)(p34;q22;q22) as recently described in our previous report [19]. However, as far as we know our study is the second to identify the involvement of 1q32 in a three-way translocation involving t(8;21). The first has been reported by Kondo et al. [20]. This suggests that 1q32 region may harbour a candidate gene critically involved in the pathogenesis of AML, as a several additional mutational event occurring in hematopoietic progenitors and blocking the normal path of differentiation. In deed, the acquisition and expression of AML1/ETO from t(8;21) in a hematopoietic stem cell has a key role in leukemic transformation, since it suppresses the function of the normal AML1 protein but it does not block normal differentiation of stem cells [21]. In the both cases including the ours with the t(21;8;1) (q22;q22;q32), the patients responded to treatment with complete remission after receiving a standard course of induction therapy suggesting that the prognosis of variant t(8;21) might not differ from those of classic t(8;21). Nevertheless, the clinical significance of the three-way translocations is controversial till now because of the limited number of cases and needs further examination. Recent data suggest that a somatic mutation of KIT, the protooncogene that encodes the tyrosine kinase receptor for stem cell factor, is closely correlated with t(8;21) leukemia. This mutation is considered as a secondary oncogenic event based on t(8;21) that may accelerate the disease process and has a negative impact on the outcome of AML with t(8;21) as demonstrated by Cairoli et al. [22]. Unfortunately, we have neither sufficient BM aspirated specimen nor peripheral blood of this patient to detect this eventual C-KIT mutation.

Conclusion

Due to the rarity of this translocation, further accumulation and genetic exploring of similar cases are needed to elucidate the clinical and molecular significance of this unusual karyotypic finding.

Conflicts of interest: none.
Current practice

References


