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Letter to the Editor

A novel specific signature of pediatric MOZ-CBP acute myeloid leukemia

The reciprocal chromosomal rearrangement t(8;16)(p11;p13) is rare both in adult and pediatric acute myeloid leukemias (AML) accounting 0.2–0.4% of de novo AML and 6.5% of the M4/M5 FAB subtype, and is associated to an extremely poor prognosis [1]. This translocation is characterized by disruption and fusion of *MYST3/MOZ* on chromosome region 8p11, which encodes for a histone acetyltransferase, to *CBP/CREBBP* on 16p13, which encodes for a transcriptional co-activator and acetyltransferase [1]. This rearrangement generates a novel MOZ-CBP fusion protein that inhibits *RUNX1* regulated transcription, leading to differentiation block. The majority of cases of AML with t(8;16) are secondary to therapy and to date it is not known whether there is some underlying genetic predisposition [2]. Here we report a case of a 10 years old girl with a *de novo* AML M5b with t(8;16) as single chromosomal abnormality. This is the first pediatric case of AML with t(8;16) where both genomic and gene expression profiles have been analyzed by high resolution techniques.

Peripheral blood analysis showed a WBC of 11,000/mmc with 23% blasts, Hb 11.8 g/dL, PLT 4.1000/mmc. The bone marrow (BM) was hyperplastic with 80% blasts of AML FAB M5b by immunophenotype and morphology. Cytomorphologic analysis of BM showed myeloperossidasis positive myeloblasts with several phenomena of erythrophagocytosis (Fig. 1A), which is often displayed by the t(8;16) AML [1].

Translocation t(8;16)(p11;p13) has been detected by cytogenetic study of BM cell culture, performed according to standard techniques for G-banding, which revealed an abnormal karyotype of 46,XX,t(8;16)(p11;p13) in all the metaphases analyzed (Fig. 1B). Cytogenetic examination identified this translocation as the only chromosomal abnormality. The genomic rearrangement of *MOZ* and *CBP* has been confirmed by fluorescence in situ hybridization (FISH) (Fig. 1C), performed using a probes for the *MOZ* gene (BAC clone RP11-313J18 labeled with Spectrum Red) and a probe that cover the *CBP* breakpoint region (RP11-75P12 labeled with Spectrum Green). To identify possible cooperating oncogenic events in leukemogenesis, apart from t(8;16) translocation, a SNP array analysis was performed on leukemic blast cells at diagnosis using Affymetrix Mapping 250K Sty chips. Copy number and LOH analyses showed the complete absence of any LOH event or cryptic gains and losses of genomic material, apart from physiological copy number variation regions (data not shown). Therefore, these analyses confirmed that t(8;16) is the sole genomic abnormality of this AML case, reinforcing the role of this fusion protein as a leukemogenic trigger.

Thus far, seven different transcript types in AML with t(8;16) have been identified by reverse transcription RT–PCR [3] and molecular analysis of our case detected the *MOZ-CBP* transcript

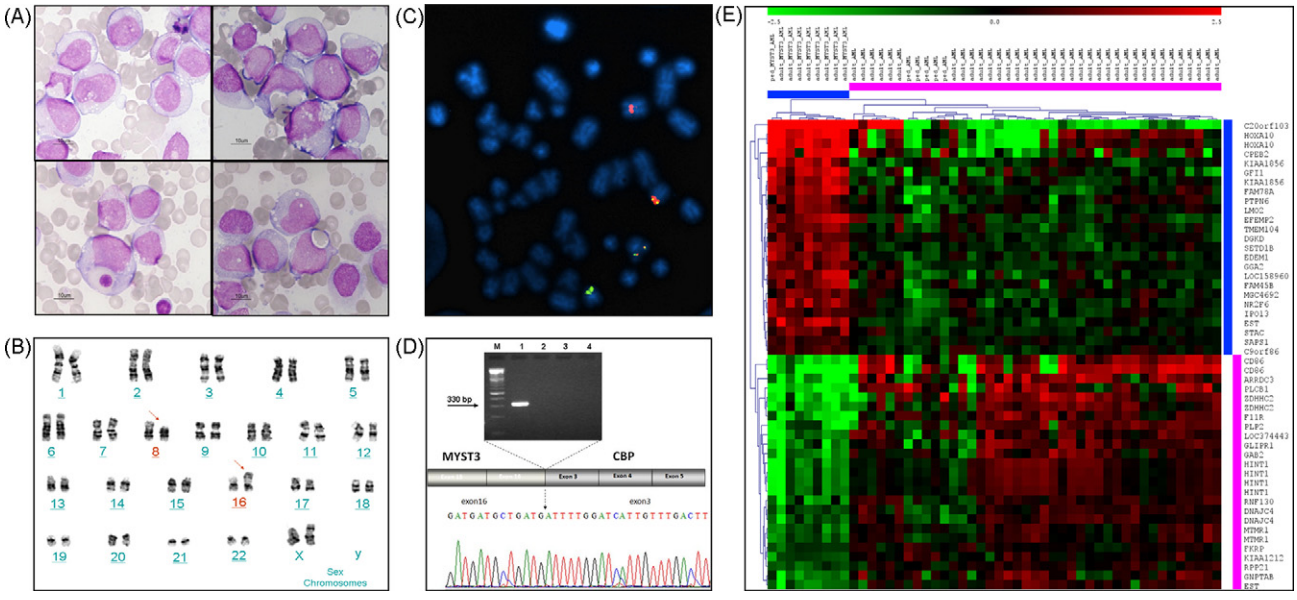

Fig. 1. (A) Patient's bone marrow smears showed erytrophagocytosis. (B) Cytogenetic analysis identified t(8;16)(p11;p13). (C) FISH analysis confirmed the translocation. (D) Chromatogram showed in-frame fusion between *MOZ* exon 16 and *CBP* exon 3. (E) Gene expression was analyzed on the pediatric case carrying the translocation (ped_MYST3_AML) on five other pediatric AML (ped_AML) and on adult AML cases from the Murati et al. paper [5].

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of type I. Actually, total RNA was extracted from bone marrow cells of the patient and cDNA was prepared. Nested RT-PCR was performed as described by Schmidt at al. [4] and the 330 bp DNA product was sequenced. Data base searches were performed using the BLAST algorithm from the National Center for Biotechnology Information (http://www.ncbi.nlm.nih.gov/BLAST/) and sequencing analysis revealed the in-frame fusion transcripts between *MOZ* exon 16 and *CBP* exon 3 (Fig. 1D).

Recently a specific gene expression signature with overexpression of *HOXA* genes and other genes regulating stem cell features and self-renewal was identified in AML with t(8;16) [5]. This manuscript analyzed the expression and copy number profile of adult and adolescent cases (M45-020 and M45-23) of AML with MYST3 translocation. We analyzed the gene expression profile of the pediatric case with t(8;16) and other five cases of pediatric AML (two M1, one M4, two M5) using Affymetrix U133Plus array. Raw data from the six pediatric patients were normalized with *rma* algorithm and merged with *rma*-normalized Affymetrix data from the dataset by Murati et al. [5]. The signature induced by t(8;16) in adult *de novo* and secondary AML with *MYST3-CREBBP* fusion transcript [5] was shared also by our pediatric case that showed, among others, the marked overexpression of *HOXA9*, *HOXA10*, *HOXA11*, *LMO2*, *PTPN6* and *GFI1* genes (Fig. 1E). Moreover we found an overexpression of *MYB*, in agreement with data on other adult cases. Many of these features are shared with leukemias harboring MLL-rearrangements or T-ALL. The results of this analysis confirms the existence of a specific *MYST3-CREBBP* leukemic signature, that is shared by adult and pediatric AML cases, characterized by the expression of genes responsible of self-renewal and stem cell features, resembling other poor prognosis groups of leukemias.

Patient received induction chemotherapy according to National Protocol for Pediatric Acute Myeloid Leukemia (AIEOP AML 2002/01) that includes two cycles ICE (Idarubicine, Cytarabine, Etoposide) [6]. A complete response of bone marrow was documented both by morphology and molecular biology/FISH at the end of first cycle. During second ICE the patient experienced fatal hepatic and hematological toxicities characterized by massive hemolysis, hyperbilirubinemia and liver enlargement that lead rapidly to death.

This case confirm that AML with t(8;16) is characterized by a poor outcome, but, even if in literature a low rate of response to chemotherapy is reported [7], our patient get rapidly an hematological remission. However the rapid evolution of toxicity did not allow her to perform allogenic transplantation, which might improve outcome in patients with AML (8:16).

Conflict of interest

All authors have no conflict of interest to report.

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Contributions. SS provided the conception and design of the study and performed cytogenetic and FISH analysis; FM collected clinical data, took care of patient and supplied the drafting of manuscript; AA performed SNP array and gene expression analysis and supplied the drafting of manuscript; VL performed molecular and sequencing analysis; RM collected clinical data and took care of patient; AP supplied the design of the study and revised the article critically for important intellectual content and gave final approval of the version to be submitted.

References

[1] Murati A, Adelaide J, Quilichini B, Remy V, Sainty D, Stoppa AM, Bernard P, Olschwang S, Birnbaum D, Chaffanet M, et al. New types of MYST3-CBP and CBP-MYST3 fusion transcripts in t(8;16)(p11;p13) acute myeloid leukemias. Haematologica 2007;92(2):262–3.

[2] Boyd EM, Bench AJ, Vaghela KJ, Campbell GN, Chowdhury FB, Gudgin EJ, Scott MA, Erber WN. Therapy-related acute myeloid leukaemia with t(8;16)(p11;p13);MOZ-CBP and polymorphisms in detoxifying and DNA repair genes. Leukemia 2009;23(6):1164–7.

[3] Terui K, Sato T, Sasaki S, Kudo K, Kamio T, Ito E. Two novel variants of MOZ-CBP fusion transcripts in spontaneously remitted infant leukemia with t(1;16;8)(p13;p13;p11), a new variant of t(8;16)(p11;p13). Haematologica 2008;93(10):1591–3.

[4] Schmidt HH, Strehl S, Thaler D, Strunk D, Sill H, Linkesch W, Jager U, Sperr W, Greinix HT, Konig M, et al. RT-PCR and FISH analysis of acute myeloid leukemia with t(8;16)(p11;p13) and chimeric MOZ and CBP transcripts: breakpoint cluster region and clinical implications. Leukemia 2004;18(6):1115–21.

[5] Murati A, Gervais C, Carbuccia N, Finetti P, Cervera N, Adelaide J, Struski S, Lippert E, Mugneret F, Tigaud I, et al. Genome profiling of acute myelomonocytic leukemia: alteration of the MYB locus in MYST3-linked cases. Leukemia 2009;23(1):85–94.

[6] Pession A, Rizzari C, Putti M, Masetti R, Casale F, Fagioli F, Lo Nigro L, Luciani M, Micalizzi C, Menna G, et al. Results of the AIEOP AML 2002/01 Study for Treatment of Children with Acute Myeloid Leukemia. Blood 2009;114(22):13.

[7] Haferlach T, Kohlmann A, Klein HU, Ruckert C, Dugas M, Williams PM, Kern W, Schnittger S, Bacher U, Loffler H, et al. AML with translocation t(8;16)(p11;p13) demonstrates unique cytomorphological, cytogenetic, molecular and prognostic features. Leukemia 2009;23(5):934–43.

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