Relato de Caso / Case Report

t(4;11) (q21;q23) in acute myeloid leukemia-M0 following treatment [EW92 Protocol] for Ewing's sarcoma

Leucemia mielóide aguda-M0 com t(4;11) (q21;q23) após tratamento para sarcoma de Ewing com o protocolo EW92

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*We report on a 7-year-old girl with Ewing's Sarcoma (ES) who developed a poorly differentiated acute myeloid leukemia (AML-M0) 20 months after beginning the EW92 protocol for the treatment of the primary tumor. She received a total dose of 1500 mg of etoposide, a tumor cumulative radiation dose of 35Gy and granulocyte colony-stimulating factor (G-CSF) was as predicted in the protocol regimen. At onset of secondary malignancy her laboratorial analysis revealed immature blast cells characterized by CD34+/CD33-/a-MPO+ and a t(4;11)(q21;q23) abnormality. This serious complication of ES treatment, which associates etoposide, irradiation and G-CSF schedule, should be weighed against its therapeutic benefits. Rev. bras. hematol. hemoter. 2004;**26**(2):122-125.*

***Key words**: Therapy-related acute myeloid leukemia,* MLL *gene, Ewing's sarcoma, Topoisomerase II inhibitors, G-CSF.*

Introduction

A great success in the treatment of malignant diseases has been obtained worldwide and children with cancer are expected to survive more than 5 years. The addition of cytokines and/or granulocyte colony-stimulating factor (G-CSF) into treatment regimens has allowed the intensification of multidrug chemotherapy and better clinical outcomes of some types of pediatric cancers.[1] However, long-term effects of childhood cancer therapy are still a problem that deserve attention mainly because of the development of secondary malignancies.[2,3] One of the most important of these late effects is secondary acute leukemia (SAL).[4,5] SAL is mainly caused by treatment with mechlorethamine, or by the use of etoposide, which is an inhibitor agent of the DNA enzyme topoisomerase II.[6,7] In any of these circumstances frequent cytogenetic abnormalities are found. They are characterized either by monosomies 5 and 7 or deletions when patients are treated by alkylanting agents, or by non-random rearrangements of 11q23 in cases treated by topoisomerase II inhibitor agents.[8]

Ewing's sarcoma (ES) belongs to a group of primitive cells originating from the neuroectoderma and are associated to t(11;22)(q29;q12).[9-11] As morphologically undifferentiated, ES is difficult to distinguish from other neuroectodermal tumors and acute leukemia. An excessive risk of secondary malignancies has been reported in survivors of ES, mainly those who received alkylanting agents and radiation in different protocols.[12,13] The new

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strategy for treatment is the use of high-doses in short-term chemotherapy and the incorporation of granulocyte colony-stimulating factor (G-CSF) to prevent severe neutropenia.[1] Here we report on a case of SAL, in which cells were characterized by a very immature sub-type of myeloid leukemia presenting a t(4;11)(q21;q23) abnormality. Due to the short period of time from the beginning of EW92 treatment, to the onset of leukemia, we suggest that G-CSF with the potential to raise the proliferation of a genetically unstable clones of damaged stem cells, may have played an important role in the development of SAL.

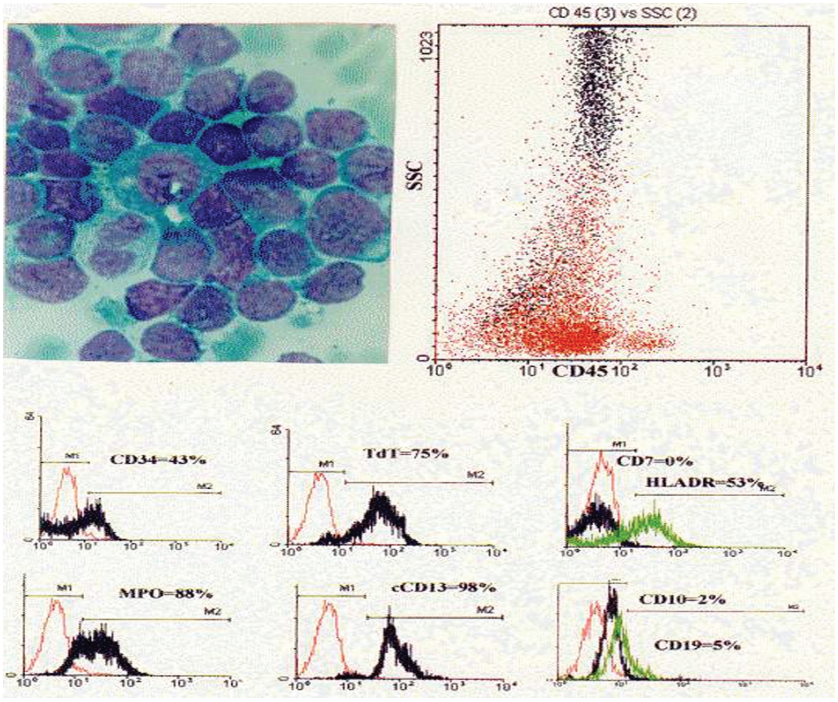

Fig. 2

Case Report

A 7-year-old girl was admitted to the Centro de Oncologia Pediatrica in Recife, Brazil in January 1999, suffering from Ewing's sarcoma of the right clavicle. At the time of admission she had no complains regarding fever, weight lost or nocturnal sweating. On physical examination she revealed a painful tumor [4 x 5 cm] beneath the right clavicle but no signs of acute inflammatory processes. Laboratory findings were as follows: Hemoglobin, 12 g/dL; white blood cell (WCC) count 5 x 10^6/mL with no abnormal cells; platelet count of 37.6.x 10^6/mL; Biochemical tests revealed serum calcium (10.9 mg/dL) and a high level of lactic dehydrogenase (1035 IU/L). Bone marrow aspirate and trephine biopsies were normal. Tumor tissue biopsy was characterized by infiltration of round cells compatible with Ewing's sarcoma (Figure 1). Clinically she was stage I (TI/N0/M0). Polychemotherapy according to the EW-92 protocol scheme [ifosfamide, etoposide, cyclophosphamide, doxorubicin, vincristine and dactinomycin] was

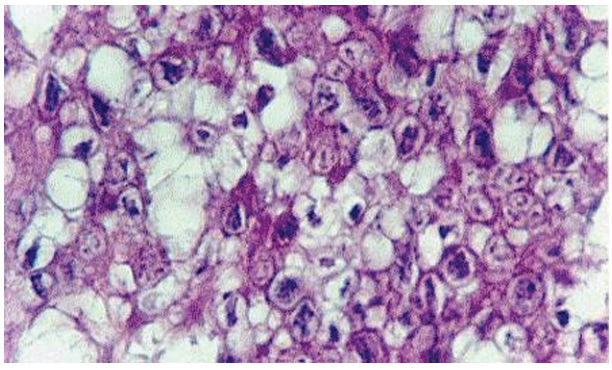

Fig. 1

Fig. 3

administered for 16 weeks, followed by local radiotherapy (total dose 35Gy). The total dose of etoposide was 1500 mg and for cyclophosphamide the total dose was 7000 mg/m.[2] She also received G-CSF (5 mg/kg/day, subcutaneous) for 5 consecutive days at each chemotherapy cycle. The cumulative dose of G-CSF was 4950 mg/kg for the complete treatment. Despite the prophylactic treatment with anti-fungal agents she had several episodes of opportunistic infections and chemotherapy was discontinued for several weeks. In October 2000, she was readmitted with fever and the WCC was 19.7x10^6/mL with 68% blast cells, platelets count 15x10^6/mL, hemoglobin, 10 g/dL. Bone marrow dry tap aspiration with infiltration by 80% of blast cells and cytological examination showed small to medium-sized cells with basophilic, scarce and agranular cytoplasm. PAS and Sudan black cytochemical reactions were negative. Immunophenotyping by flow

cytometry displayed CD45=88%, CD34=43%, TdT=75, HLADr=53%, cCD3=4%, CD7=0%, CD10=2%, CD19=5%, CD33=4%, CD13= 5%, cCD13=98%, CD14=0%, a-MPO=88%, compatible with immature myeloid leukemia, FAB M0-subtype (Figure 2 a and b). Cells were cultured in RPMI with 20% of fetal calf serum for 72 hours with phytohemaglutinin stimulation. GTG banding was performed and chromosomes were analyzed according to International System of Human Cytogenetics.[14] Cytogenetic analyses showed a karyotype characterized by 46,XX, t (4;11)(q23;q23), in 11 mitosis obtained as shown in Figure 3.

Polychemotherapy with cyrosine-arabinoside, iadurubicin and deoxyrubicin was initiated according to BFM-95 scheme. She had a good clinical response and so far, she is still in remission 60 months following from the diagnosis of ES.

Discussion

It is well accepted that chemotherapy combined with radiotherapy given in extended fields added to genetic factors could be associated with the development of SAL. Recently, Relling et al. described the risk of secondary myeloid malignancy after etoposide treatment in twenty children out of 412 children treated on ALL protocols. In the setting of intensive antileukemic therapy, short-term use of G-CSF had an increase risk of SAL (7.2%) when compared with children who received either irradiation or G-CSF (1.3%). The cumulative incidence functions differed significantly (p=0.017).[18]

Our patient had an ES, which was treated with the EW92 protocol that includes etoposide associated with cyclophosphamide, local radiotherapy and G-CSF.[1] The clinical and laboratorial features in this case are typical of secondary leukemia after treatment with topoisomerase II inhibitor agents. There was no evidence of myelodysplasia previously and the interval from the beginning of previous treatment to the onset of leukemia was only 20 months. Intriguing was the fact that she had <1500 mg/m^2 as the total etoposide dose when the development of secondary leukemias is closely related to >2000mg/m^2 given twice a week.[7] Another unexpected finding was the t(4;11) (q23;q23) abnormality in an AML-M0. This translocation has been frequently reported in pro-B ALL or in AML-M4/M5 types, and present anomalies at band 11q23 are caused by drugs that inhibit topoisomerase II. So far, to the best of our knowledge, no case has been reported with AML-M0 with t(4;11) after treatment with topoisomerase II inhibitors. In addition, recent reports show that the inclusion of granulocyte colony stimulating factors in treatment with anti neoplastic drugs may lead to medullar dysplasia and leukemia.[1] Our hypothesis in this case is that high dose alkylating and topoisomerase-II inhibitors caused genetic damage in the myeloid-committed stem cell translated by t(4;11) and this clone was offset by the rapid turnover of stem cells induced by the frequent use of G-CSF. Therefore, it is important to evaluate this observation when multidrug chemotherapy is required to treat mesenchymal tumors. The use of G-CSF in such cases should be indicated with parsimony.

Resumo

*Nós descrevemos o caso clínico de uma criança do sexo feminino, com 7 anos de idade, portadora de sarcoma de Ewing, que evoluiu com leucemia aguda mielóide pouco diferenciada (LMA-M0) após vinte meses de tratamento utilizando o protocolo EW92. Ela recebeu uma dose total de 1.500 mg de etoposídio, irradiação tumoral na dose total de 35G, e fator de estimulação de colônia granulocítica (G-CSF) conforme programação do protocolo terapêutico. Os exames laboratoriais, por ocasião do diagnóstico da segunda malignidade, mostraram células blásticas imaturas caracterizadas pela expressão de CD34+/ CD33-/aMPO+ e a translocação t(4;11) (q 21;q23). A exclusão do G-CSF nos esquemas terapêuticos que associam etoposídio e irradiação tumoral se justifica devido a esta séria complicação no tratamento do sarcoma de Ewing. Rev. bras. hematol. hemoter. 2004;**26**(2):122-125.*

Palavras-chave: *Leucemia mielóide aguda secundaária; translocação envolvendo a região 11q23 após uso de etoposídio.*

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Avaliação: Editor e dois revisores externos.
Conflito de interesse: não declarado

Recebido: 30/04/2004
Aceito: 10/06/2004