Cancer Genetics 209 (2016) 166–170

Cancer Genetics

A der(11)t(4;11)(q21;p15) in a T-ALL/LBL patient

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Translocation t(4;11)(q21;p15) is a rare recurrent change associated to T-cell acute leukemia. In most cases, this alteration appears as the only abnormality or as part of a simple karyotype. In this report, we present the first case of T acute lymphoblastic leukemia/lymphoma (T-ALL/LBL) with the unbalanced translocation der(11)t(4;11)(q21;p15) as part of a very complex karyotype with multiple chromosome abnormalities, most of them not previously described in the literature. FISH (fluorescence in situ hybridization) and spectral karyotype (HiSKY) analysis confirmed the presence of complex alterations. The patient, a 16-year-old male, showed poor response to treatment and short survival (11 months). A detailed review of previously reported cases with t(4;11)(q21;p15) is also provided. The description of this type of alterations may contribute to the identification of new molecular mechanism associated to neoplastic development.

Keywords T-cell acute leukemia/lymphoma, cytogenetics, FISH, spectral karyotype

Introduction

T-cell acute lymphoblastic leukemia/lymphoblastic lymphoma (T-ALL/LBL) is a neoplasm committed to the T-cell lineage that can be observed involving bone marrow (BM) and peripheral blood (T-acute lymphoblastic leukemia, T-ALL) or presenting with primary involvement of thymus, nodal or extranodal sites (T-lymphoblastic lymphoma, T-LBL) (1). By convention, the term lymphoma is used when the process is confined to a mass lesion with no or minimal evidence of peripheral blood and BM involvement. If the patient presents with a mass lesion and lymphoblasts in the BM, the distinction between leukemia and lymphoma is arbitrary. T-ALL comprises about 15% of childhood ALL while T-LBL correspond to about 85–90% of all lymphoblastic lymphomas; both are most frequent in male adolescents. The annual incidence is 1.6/100,000 individuals in the general population (1). In Argentina, data of the Onco-Hematology Argentine Registry (2) show an incidence of 3.0/100,000 in children under 15 years.

Cytogenetic studies in this pathology have demonstrated an abnormal karyotype in about 50% of patients (3). Thirty five percent of cases show translocations involving T cell receptors and different genes (4,5). These translocations usually result in oncogenes becoming juxtaposed to the promoter and enhancer elements of *TCR* genes, leading to their aberrant expression and the development of T-ALL/LBL. Alternatively, aberrant expression of one or more transcription factors is a critical component of the molecular pathogenesis of T-ALL/LBL (5). On the contrary, the presence of arecurrent reciprocal translocation without involvement of at least one *TCR* encoding gene is a rare event in this pathology. The translocation t(4;11)(q21;p15) is a very uncommon rearrangement described in 1985 (6) that, at the molecular level, induces the fusion of the *RAP1GDS1* (RAP1, GTP-GDP dissociation stimulator 1) gene located at band 4q21, with the *NUP98* (nucleoporin 98 kDa) gene mapped at 11p15 (7,8), leading to a new chimeric transcript encoding a protein with dominant oncogenic properties: *NUP98-RAP1GDS1* (7). In most cases, this alteration appears as the only abnormality or as part of a simple karyotype (9). The literature shows only two cases with complex karyotype (three or more alterations) (10,11) and only one case with a complex translocation (12). In this study, we present the first case of T-ALL/LBL with an unbalanced translocation der(11)t(4;11)(q21;p15) as part of a very complex karyotype

Received September 21, 2015; received in revised form December 2, 2015; accepted January 6, 2016.

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2210-7762/$ - see front matter © 2016 Elsevier Inc. All rights reserved.
http://dx.doi.org/10.1016/j.cancergen.2016.01.001

with multiple chromosome abnormalities, most of them not previously described in the literature.

Case report

A 16-year-old male was referred to the Hospital of High Complexity "Presidente Juan Domingo Perón", Formosa, Argentina, in November 2013 with cervical and inguinal lymphadenopathies and splenomegaly. Hematological data showed: white blood cells count 20×10^9/L (28% neutrophils, 62% lymphocytes, 4% monocytes, 6% eosinophils), hemoglobin 15.4 g/dL, hematocrit 44% and platelet count 138×10^9/L. Lactate dehydrogenase was raised to 448 IU/L (normal range 240–480 IU/L). The patient had a normal liver and kidney function and, negative serology for hepatitis B and C. The histopathological study of the lymph node showed complete replacement for a lymphoid population with diffuse pattern, composed of medium sized blasts with scant cytoplasm, condensed nuclear chromatin and indistinct nucleoli. Immunohistochemical analysis showed co-expression of CD1a, Tdt, CD34 and CD10, and moderate reactivity of CD3 (Figure 1a). The Ki67 antigen disclosed 80% of proliferating cells. The BM aspirate showed 90% lymphocytes with diffuse pattern, with typical T-ALL/LBL morphology. Flow cytometry showed proliferation of T-cell precursors, which accounted for 76.21% of cell population. The patient started chemotherapy according to the protocol of the Argentine Group of Acute Leukemia Treatment (GATLA) (13). A complete hematological remission was achieved. Four months after consolidation, the patient showed hematological relapse and died three months later.

Methods

Cytogenetic analysis

Cytogenetic study was performed on BM cells cultured in RPMI 1640 medium supplemented with 20% fetal calf serum (Gibco) during 24 h at 37 °C. G-banding technique was used. Chromosome abnormalities were described according to the International System for Human Cytogenetic Nomenclature (ISCN) (14).

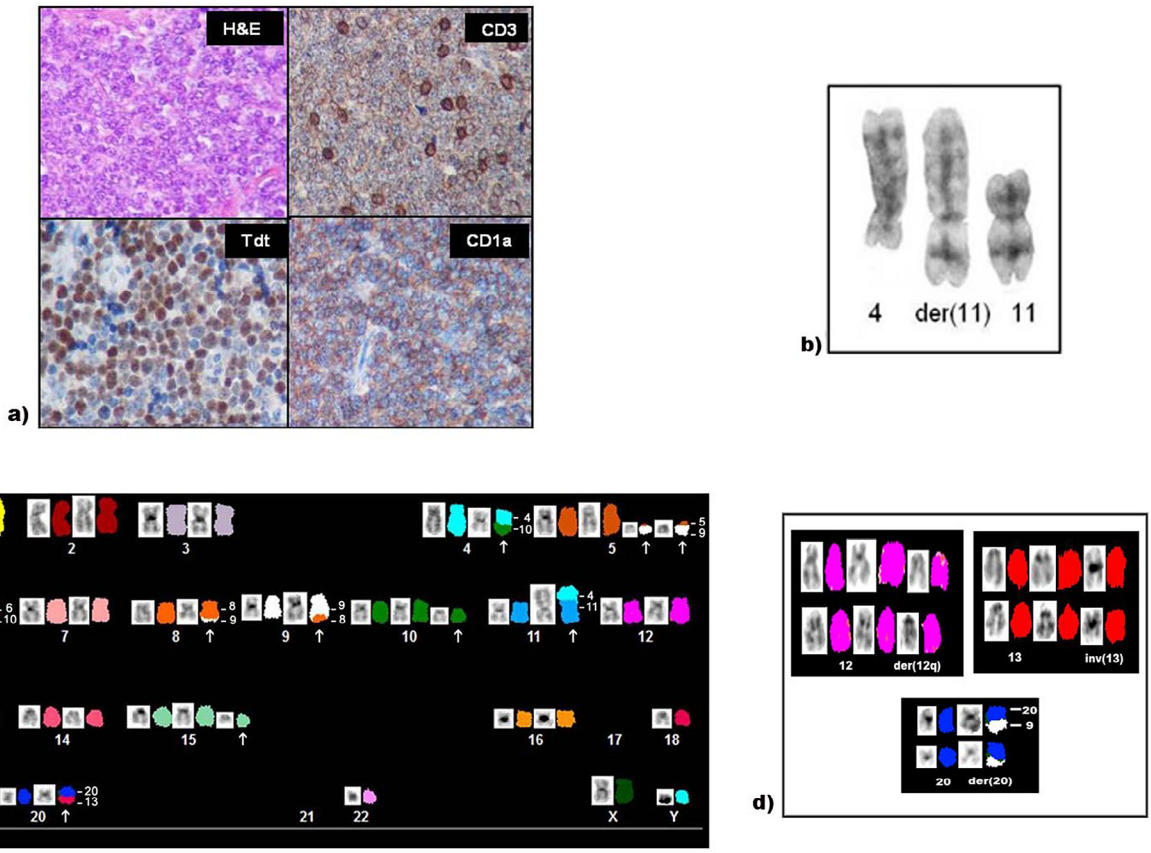

Figure 1 (a) Lymph node biopsy stained with Hematoxylin & Eosin showing complete replacement by lymphoblastic lymphoma, and immunohistochemical technique showing: moderate reactivity of CD3; Tdt expression; CD1a expression (400x); (b) G-banding partial karyotype showing normal chromosomes 4 and 11 and der(11)t(4,11)(q21;p15); (c) Spectral karyotype of the patient showing chromosome alterations: der(4)t(4;10)(q12;q?22), der(5)t(5;9)(?;?)x2, der(6)t(6;10)(q13;?), t(8;9) (q24;q34), der(10)del(10)(p11)del(10)(q24), der(11)t(4;11)(q21;p15), del(15)(q15), der(20)t(13;20) (?;q11) (arrows), as well as losses of chromosomes 17, 18, 21 and 22.The presence of a normal chromosome 9 in this cell is nonclonal; (d) G-banding and Spectral partial karyotypes showing: normal chromosomes 12 and del(12)(q11q13), normal chromosomes 13 and inv(13)(p11q14) and normal chromosome 20 and der(20)t(9;20)(?;q13).

FISH (fluorescence *in situ* hybridization) studies

FISH analysis was performed on the same material used for cytogenetic studies according to manufacturer's protocol. Total chromosome paint (TCP) FISH probes (LiVE-Lexel, Buenos Aires, Argentina) for chromosomes 4, 11 and 12 as well as LSI MLL Dual color, Break Apart rearrangement probe (11q23) (Vysis-Abbott, Illinois, USA) were used. Image acquisition was performed using Cytovision 3.9 Software (Applied Imaging Corporation, California, USA). In addition, High Resolution Spectral Karyotype (HiSKY) was performed using the ASI SKYPaint-KIT (Applied Spectral Imaging) and the BX61 HiSKY system (GenASIs Hyperspectral Platform) was employed for image acquisition.

Results

At diagnosis, cytogenetic study on the BM cells showed a 44~46,XY, der(11)t(4;11)(q21;p15),add(6)(q21),add(8)(q24),+del (12)(q11q13),–17,–19,add(20)(q11), +mar[cp9]/46,XY (10) karyotype. TCP FISH probes for chromosomes 4 and 11 confirmed the der(11)t(4;11)(q21;p15) (Figure 1b) as well as TCP12 confirmed the interstitial deletion of chromosome 12: del(12)(q11q13). The analysis with the MLL probe showed a normal pattern of signals. In order to complete the characterization of this complex karyotype, we performed HiSKY technique. The combination of G-banding analysis, FISH and HiSKY allowedrefinement of the karyotype to: 44~46,XY,der(4)t(4;10)(q12; q?22),+der(5)t(5;9)(?;?)x2, der(6)t(6;10)(q13;?), t(8;9)(q24;q34),–9,der(10)del (10)(p11)del (10)(q24),der(11)t(4;11)(q21;p15),+del(12) (q11q13),+inv(13) (p11q14),+del(15)(q15),–17,–19,der(20)t(13;20)(?;q11),der(20)t (9;20) (?;q13),–21,–21[cp17]/46,XY (11) (Figures 1c–d). In addition, the following chromosome abnormalities were observed in only one cell each: der(2)t(2;10)(q11;?), der(10)t(1;10)(?;q11), del(14)(q24). Among these alterations, only t(8;9)(q24;q34) (15) and der(20)t(13;20)(?;q11) (16) have been described in one patient each, del(15)(q15) were observed in five cases as part of complex karyotypes (9), meanwhile monosomies 9, 17 and 19 are frequent numerical alterations in T-ALL/LBL patients (9).

Discussion

The translocation t(4;11)(q21;p15) is a very infrequent rearrangement. This anomaly characterizes a subset of T-ALL/LBL originating from an early precursor with variable co-expression of CD10 and myeloid markers. It is found mainly in patients older than 15 years, and associated with poor clinical outcome (17). The translocation t(4;11)(q21;p15) may be a unique, recurring abnormality, as it appears to be associated with *de novo* acute leukemia (myeloid and lymphoid), secondary leukemias, as well as lymphoma. As mentioned, this translocation results in the fusion of *NUP98 and RAP1GDS1* genes. *NUP98* codes for a component of the nuclear pore complex (18) and is involved in numerous translocations described in hematological disorders including acute and chronic myeloid leukemias, T-cell ALL, myelodysplastic syndrome and secondary acute myeloid leukemia, suggesting an important role in leukemogenesis (19–22). *NUP98* has been found rearranged with 19 different partner genes subclassified in three different groups, with different distribution among pathologies (20). Thus, *HOX* and *NSD* genes are mainly associated with *de novo* myeloid malignancies; TOP family genes and *DDX10* are related to therapy-associated myeloid disorders, and *RAP1GDS* gene is involved in T-ALL, with the NUP98/RAP1GDS1 fusion representing 9% of the *NUP98* fusion genes. As regards to *RAP1GDS* gene, it encodes a cytoplasmic protein that regulates actin assembly associated with the folding of the membrane. When the rearrangement with *NUP98* gene occurs, the entire coding region of *RAP1GDS* merged to the GLFG repeat domain of *NUP98* to form the fusion transcript NUP98-RAP1GDS1 (7).

At present, there are 20 cases reported in the literature (Table 1): 12 patients with T-ALL, 2 with T-ALL/LBL, 2 with T-LBL, 1 with B-ALL, and 3 with acute myeloid leukemia. Six out of 16 (37.5%) cases with T-cell disease were children. Among them, eight patients showed the t(4;11) as the only karyotypic abnormality, in one of them as a result of a complex translocation, four cases had deletions of chromosomes 5, 12 or 13, one case had a different balanced translocation in the karyotype, three cases showed hyperdiploid karyotypes. From the three cases with diagnosis of AML, two had the t(4;11) as a single alteration and one case showed a hyperdiploid karyotype with multiple numerical alterations and the t(4;11)(q21;q23) involving the MLL gene. Thus, our case is, to our knowledge, the first description of this anomaly as an unbalanced translocation in the context of a very complex karyotype. In addition, complex rearrangements were observed in single cells. As known, changes in gene dose can potentially generate copy-number alterations as well as critical combinations of deleted and gained regions that are important drivers of cancer, sometimes associated to complex karyotypes (30). This remodeling of the genome may confer significant selective advantages to clonal cells that can promote cancer development and progression, associated to adverse clinical behavior of the disease. It is consistent with those observed in our patient that showed a poor clinical outcome with a very short survival.

Certainly the description of this type of alterations may contribute to the knowledge of the biological and clinical characteristics of different subgroups of patients. It is also interesting to note the importance of using molecular cytogenetic techniques in cases with complex chromosome rearrangements to better characterize the karyotype. It constitutes a contribution to a higher accuracy in the diagnosis and/or prognosis of patients and to guide future studies that enable the identification of new molecular mechanisms associated with neoplastic development that may be the basis for new therapeutic approaches.

Acknowledgments

This work was supported by grants from the National Research Council (CONICET), PIP 112 201 101 00517 the National Agency of Scientific and Technical Promotion (ANPCyT) PICT 2014-1566, and the National Cancer Institute from Argentina. The authors want to thank the technical assistance of the National Center of Medical Genetics for spectral analysis of chromosome alterations and Dr Alejandro Laudicina that kindly provided some of the FISH probes used in this study.

Table 1 Cases with t(4;11)(q21;p15) reported in the literature

Reference	Age/sex (years)	Karyotype	Diagnosis	Methodology
Hussey et al. (7)	49/M	46,XY,t(4;11)(q21;p15),del(5)(q13q31)	T-ALL	Cytog, PCR
	25/F	46,XX,t(4;11)(q21p14-15),del(12)(p13),del(13)(q12q14)	T-ALL	Cytog, PCR
Inoue et al. (6)	14/M	46,XY, t(4;11)(q21p14-15),12p-/46,XY, t(4;11)(q21p14-15)	T-ALL	Cytog
Kawakami et al. (23)	51/F	46,XX,t(4;11)(q21;p15)	T-ALL /LBL	Cytog
Hardingham et al. (24)	21/M	48,XY,t(4;11)(q21;p15),+2mar	T-ALL	Cytog
Pui et al. (25)	6/F	46,XY, t(4;11)(q21;p14-15)	T-ALL	Cytog
Bloomfield et al. (26)	40/M	46,XY, t(4;11)(q21;p15)	T-ALL	Cytog
Mecucci et al. (8)	16/F	47,XX,t(4;11)(q21;p15),+8	T-ALL	Cytog, FISH
	38/F	46-47,XX,t(4;11)(q21;p15),+mar	T-ALL	Cytog, PCR?
Thangavelu et al. (27)	11/M	46,XY,t(4;11)(q21;p15)	T-LBL	Cytog, FISH
	36/M	46,XY,t(4;11)(q21;p15)	AML	Cytog, FISH
Douet-Guilbert et al. (12)	40/F	46,XX,t(4;11)(q21;p15)/46,XX	T-ALL	Cytog, FISH
	25/M	46,XY,t(1;4;11)(p32;q21;p15)/46,XY	T-ALL	Cytog, FISH
Kobzev et al. (28)	18/M	46XX,t(2;21)(q11;q11),t(4;11)(q21;p15)/46,XX	T-LBL	FISH
Romana et al. (20)	19/M	46,XY,t(4;11)(q21;p15)	T-ALL	Cytog, FISH, PCR
	10/F	46,XY,t(4;11)(q21;p15)	T-ALL	
Yasar et al. (19)	32/M	46,XY,t(4;11)(q21;q23)/46,XY,t(4;11)(q21;p15)	B-ALL	CGH array
Zhang et al. (11)	12/F	46,XX,t(4;11)(q21;p15),del(5)(q22q35),del(12)(p12)	T-ALL /LBL	Cytog, PCR
Radtke et al. (29)	?/F	46,XX,t(4;11)(q21;p15)	AML	Cytog
Secker-Walker et al. (10)	1/F	38~53,XX,+4,t(4;11)(q21;p15),t(4;11)(q21;q23),+6,–8,–9, –10,+11,–18,+19	AML	Cytog
This report	16/M	44~46,XY,der(4)t(4;10)(q12;**q?22**),+der(5)t(5;9)(?;?)x2, der(6)t(6;10)(q13;?),t(8;9)(q24;q34),–9,der(10)del**(10)** (p11)del**(10)**(q24),der(11)t(4;11)(q21;p15),+del(12) (q11q13),+inv(13)(p11q14), +del(15)(q15),–17, –19, der(20)t(13;20) (?;q11),der(20)t(9;20)(?;q13),–21**,–21** [cp17]/ 46,XY (11)	T-ALL /LBL	Cytog, FISH, HiSky

Abbreviations: M, male; F, female; Cytog, cytogenetics; PCR, polymerase chain reaction.

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