Primary cutaneous marginal zone B-cell lymphoma with prominent T-cell component and aberrant dual (T and B) genotype

Diagnostic usefulness of laser capture microdissection

Fernando Gallardo, MD1, Ramon M. Pujol, MD2, Beatriz Bellosillo, PhD2, Dolors Ferrer, MD3, Susana Boluda, MD3, Carlos Barranco, MD3, Mercè Planagumà, MD4, and Sergi Serrano, MD3.

1Department of Dermatology, 2Laboratory of Cytogenetics and Molecular Biology. 3Department of Pathology. Hospital del Mar. IMAS. Barcelona. Hospital de Palamós, Girona, Spain

Introduction

The presence of a dominant B- or T-cell clone is an important diagnostic criterion for distinguishing cutaneous lymphomas from lymphoid reactive infiltrates. Rarely, a combined B- and T-cell rearrangement can be detected from a single sample. In such instances, genotypic analysis does not permit to differentiate the presence of a single clone harbouring a mononclonal rearrangement for both IgH and TCR genes (aberrant or dual genotype) from the coexistence of both T- and B-cell monoclonal population. We herein report a patient with a dense cutaneous lymphoid infiltrate with a double prominent B- and T-cell component. A dual B- and T-cell clonality by PCR was detected from DNA extracted from the entire biopsy. Genotypic analysis with PCR obtained after laser assisted microdissection from the B-cell population, showed again both T- and B-cell monoclonal rearrangements. Conversely, the microdissected T-cell population did not reveal a monoclonal pattern. The diagnosis of cutaneous B-cell lymphoma with a dual B- and T-cell genotype was established.

Case report

A 68-year-old woman presenting a 6 month solitary nodule on her upper back was referred to our department. Past medical history was uneventful. Physical examination revealed an apparently healthy woman presenting a well-defined, firm, dome-shaped nodule on her upper back (Fig. 1). No similar lesions were noted elsewhere. Neither enlarged regional lymph nodes nor hepatosplenomegaly were present. The patient presented no B symptoms.

Results

Histopathological and immunohistochemical study

Clonality studies

DNA obtained from two 15 micrometer sections of paraffin embedded biopsies using the QiAamp Tissue Kit (QiAGEN GmbH, Hilden, Germany) was amplified by PCR with consensus primers for the TCR gamma chain (IgH: monoclonal) (TCR: monoclonal) and IgH (IgH: germline) (TCR: polyclonal) (IgH: monoclonal) (TCR: monoclonal).

Laser capture microdissection (LCM)

Approximately 600 CD20 and CD3 positive cells were captured using P.A.L.M. Microlaser (Bensried, Munchen, Germany) and single cells were transferred into a sterile microtube by a laser pressure catapulting system. After digestion overnight with protease K at 55ºC the extract from both samples was used directly to study the IgH and TCR genes rearrangement by PCR.

Comment

The presence of the so-called aberrant or dual rearrangement (Lineage infidelity) can be detected in up to 5-20% of cases of both mature T- or B-cell lymphomas. Some observations seem to indicate that the prevalence of a dual genotype occurs in a higher proportion (up to 50%) of Sézary’s syndrome cases. However, aberrant dual genotype resulting from a lineage infidelity has rarely been reported in cutaneous lymphomas. Our case also illustrates the diagnostic difficulties that may represent B-cell lymphomas presenting a prominent T-cell reactive population, based exclusively on morphologic and immunophenotypic features. In MALT lymphomas, the presence of an accompanying reactive polyclonal T-cell population is an almost constant phenomenon. Rarely, these non-neoplastic accompanying T-cells may be so intense that obscures the B-cell neoplastic population or can show a clonal or oligoclonal gene rearrangement.

Laser-assisted microdissection allowed us to demonstrate a dual genotype in the B-cell component of the lesion, whereas T-cell areas showed a polyclonal pattern. The combination of clinical, histopathological and immunophenotypic data and genotypic features on microdissected cells enabled us to establish the definite diagnosis of PCBCL (marginal zone type).

References


Acknowledgments

This study was partially supported by the grants FIS 00/225, FIS 01/1424, FIS 02/0022, Xarxa Temàtica de Limfomas Cutànis de la Generalitat de Catalunya 2002/XT/00020 and 003/179 from the Spanish “Ministerio de Sanidad y Consumo”.

Methods

Histopathological and immunohistochemical study

The primary antibodies used were: CD3(clone F 7.2.38), CD6(C8/144B), CD43(DF71), CD20(L26), CD79a(JCB 117), bcl6(PG-B6p), CD30(Ber-H2), ki67(MIB-1), kappa and lambda (DakoCytomation, Glostrup, Denmark); CD1D2P2D1F11, CD4(1F6), CD7(272), CD5(4C7), CD23(1B-12), CD56(1B-6) and p53(D07) (Novocastra, Newcastle, UK); TIA-1 (Immunotech, Marseille, France) and BCL2(clooney 100) (BioGenex, San Ramon, CA, USA). The detection system employed was En Vision Plus (DakoCytomation).

Laser-assisted microdissection from DNA extracted from the entire biopsy. Genotypic analysis with PCR obtained after laser assisted microdissection from the B-cell population, showed again both T- and B-cell monoclonal rearrangements. Conversely, the microdissected T-cell population did not reveal a monoclonal pattern. The diagnosis of cutaneous B-cell lymphoma with a dual B- and T-cell genotype was established.