

CD20, CD5, CD10, BCL-2, BCL-6, c-MYC e para a proteína Ki-67 (> 90%), um marcador biológico que indica proliferação tumoral. Nesta técnica, o marcador CD45 também foi negativo para as células linfóides neoplásicas. A pesquisa de rearranjo dos genes c-MYC e BCL-2 por FISH foi positiva nesta amostra. O estudo medular foi negativo para infiltração neoplásica. **Discussão:** Os linfomas não Hodgkin B (LNH-B) são um grupo complexo e heterogêneo de neoplasia, composto por mais de 80 tipos distintos. Entre esses grupos, podemos destacar o LNH-B de alto grau, um subgrupo agressivo e raro do Linfoma Difuso de Grandes Células B (LDGCB), que compartilha características moleculares intermediárias entre o LDGCB e Linfoma de Burkitt (LB). Normalmente, na morfologia estes tumores apresentam grau intermediário ou alto. A expressão c-MYC pode ocorrer em 30 a 60% dos casos, quando apresentam expressão de c-MYC e BCL2 ou BCL6 são classificados como “double hit” ou “triple hit” quando apresentam expressão de c-MYC, BCL2 e BCL6. O CD45 é universalmente expresso (variável intensidade) nas células nucleadas hematopoiéticas normais. Os LNH-B expressam tipicamente o marcador pan-leucocitário CD45 e a perda da expressão deste marcador é rara. Não foi evidenciada expressão positiva para o marcador CD45 (clone J33) através da análise imunofenotípica. Além disso, a expressão de CD45 foi normal nas demais populações leucocitárias evidenciando a boa performance do anticorpo utilizado. Este achado foi confirmado pelos resultados imunohistoquímicos, que mostrou CD45 (LCA) (clone 2B11+PD7/26) negativo na população de células linfóides neoplásicas. O que também torna esse resultado mais robusto é que a avaliação do CD45 foi testada com diferentes clones de anticorpos monoclonais. **Conclusão:** O presente relato ressalta a raridade de um caso de Linfoma não Hodgkin B de alto grau “double hit” com ausência da expressão de CD45. Na literatura há raros casos desta ocorrência em Linfoma Folicular e Linfoma extra nodal, mas não encontramos nenhum relato de LNH de alto grau “double hit” na literatura.

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USE OF TCR V β 2 REPERTOIRES TO DIFFERENTIATE CD8 T-CELL LYMPHOMAS FROM INFECTIOUS DISEASE REACTIVE LYMPHOCYTOSIS

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Introduction: Reactive lymphocytosis can be difficult to differentiate from clonal lymphocytosis, especially when it involves CD8 positive T-lymphocytes. Leukemias and lymphomas cause an increase in this population at the expense of replication of clonal cells with genetic and/or phenotypic aberrations. Infections, mainly of viral etiology, often cause an exacerbated response of this population. The expansion resulting from a viral infection is not selective, so multiple

cell clones are involved. Defining the cause of lymphocytosis is essential for determining therapeutic measures. **Purpose:** To define T-cell clonality in patients with peripheral blood lymphocytosis using TCR V β 2 repertoire by flow cytometry multiparametric analysis (FCM). **Materials and methods:** We included five patients with suspected T-cell lymphocytosis, one patient with immunodeficiency, and one control. Analysis of TCR V β 2 rearrangements was performed on whole blood sample using the IOTest Beta Mark TCR V β Repertoire Kit (IOTest, Beckman Coulter, Indianapolis, USA) antibody set. BD FACSCanto™ II Cytometer and Infinicyt™ 2.0 analysis software were used. **Results:** Patient 1 was considered as negative control (lymphocytes $3.41 \times 10^3/\mu\text{L}$) showed expression of various V β 2 clones, ranging from 0.05 to 1.40%. Patients 2 and 3, both increased CD8 T-lymphocytes associated with a history of infectious disease, had a polyclonal profile of TCR V β 2 expression, Patient 4 had lymphocytosis ($4.25 \times 10^3/\mu\text{L}$) with majority expression of V β 5.1 clones on CD4 (10.84%) and CD8 (10.56%) lymphocytes and of V β 13.2 clone on CD4 lymphocytes (11.53%). This patient was considered with reactive oligoclonal proliferation. Patient 5 had lymphocytosis with an abnormal CD8 T cell phenotype suggestive of Large Granular T-Cell Leukemia. TCR rearrangement analysis confirmed the presence of the V β 13.1 clone in CD8 T-cells. Patient 6 had a large lymphocytosis ($462.92 \times 10^3/\mu\text{L}$) with a predominance of CD8 positive T-cells and a phenotype suggestive of Prolymphocytic T Cell Leukemia. TCR rearrangement indicated the majority presence of the V β 8 clone on CD8 T-cell. And finally, patient 7 had mild lymphocytosis ($6.87 \times 10^3/\mu\text{L}$) with a predominance of CD8 T-cells with expression of clone V β 4 (8.90%) and V β 17 (2.01%) and of clone V β 3 on CD4 cells (6.51%) and was considered as oligoclonal expansion. **Discussion:** The confirmation of monoclonal expansion of CD8 T lymphocytes could help in the diagnosis of T-cell neoplasms, confirming the diagnosis of large granular T-cell leukemia and CD8 T-cell prolymphocytic leukemia in two patients. Among the patients with polyclonal or oligoclonal expression, all patients had a history of viral infectious diseases, such as CMV or EBV infection. Patient 7 had a diagnosis of immunodeficiency with oligoclonal T expansion that can be discussed separately. **Conclusion:** Patients presenting with CD8 positive T lymphocytosis and often nonspecific clinical symptoms may benefit from combined evaluation of phenotype and clonal expression by the V β 2 repertoire.

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ENCONTROS VIRTUAIS NA EDUCAÇÃO CONTINUADA

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A pandemia do COVID-19 trouxe diversos obstáculos, mudando as rotinas de vida social e profissional, nos afastando fisicamente. No entanto, sabendo da importância de

