or bone marrow samples (4mL), nucleic acids were stabilized and then extracted using Magna 96 (Roche). A one-step RT-qPCR QuantiNova Probe master mix (Qiagen) with primers and probes described by Gabert et al. 2003 (EAC) and by Pane et al. 1996 were used. BCR-ABL1 and ABL RNAs were co-amplified at Roche LightCycler 480II for e14a2/e13a2 (p210), e1a2 (p190), and e19a2 (p230) fusions. The degree of agreement between the test methods (flow cytometry) and the comparative methods (PCR for BCR-ABL) was quantified using Kappa statistics with three categories. **Results:** In this period, 21 samples from different patients were received at Sabin Medicina Diagnostica lab for CML investigation and medical order for flow cytometry and BCR-ABL assays. 10 samples were from peripheral blood and 11 from bone marrow. In 10 samples (3 bone marrows and 7 peripheral blood), flow cytometry assay did not show a CD26+ CML LSC population, and BCR/ABL PCR assay resulted negative. In 11 samples (8 bone marrows and 3 peripheral blood), a CD26+ CML LSC population was identified by flow cytometry and BCR-ABL PCR assay resulted positive. There were no discordant results. The degree of agreement between the test methods (flow cytometry) and the comparative methods (PCR for BCR-ABL) was a perfect agreement (kappa=1). **Conclusions:** In conclusion, our data are in accordance with the results previously described by Raspadori et al. Although we still believe that further studies are necessary, the identification of a CD26+ CML LSC population by flow cytometry may be a diagnostic tool for CML when a BCR-ABL PCR assay is not available.

https://doi.org/10.1016/j.htct.2020.10.746

745

CYTOLOGICAL AND MULTIPARAMETRIC FLOW CYTOMETRY ANALYSIS FOR DIAGNOSTIC OF BREAST IMPLANT-ASSOCIATED ANAPLASTIC LARGE CELL LYMPHOMA

A.P. Azambuja, F. Gevert, R.O. Merhy, A.P. Sebastiao, A.C. Groth

Hospital Nossa Senhora das Graças, Rio de Janeiro, RJ, Brazil

Background: Breast implant-associated anaplastic large cell lymphoma (BIA-ALCL) is a provisional entity with morphological and immunophenotypic characteristics indistinguishable from anaplastic large cell lymphoma (ALCL), as hallmark morphology and CD30 positivity. However, unlike ALCL, BIA-ALCL often presents as unilateral effusion associated to silicone breast implants. Diagnostic confirmation of BIA-ALCL can be difficult. In this setting, multiparametric flow cytometry (MFC) looking for CD30, HLA-DR and CD25 positivity may be a good option for help in diagnostic assistance. **Objective:** To describe cytological and flow cytometric founds of patients with suspected periprosthetic fluid and compare confirmed BIA-ALCL to negative patients. Methods: From mar/2018 and Jul/2020, all periprosthetic fluid (PF) collection sent to our lab to cytology and MFC analysis to quantification and characterization of pathologic, T and B cells were included. All specimens were collected in dry tubes and sent immediately to the lab. A cytospin was prepared and cored with Wright-Giemsa staining for morphological evaluation. A total of 100 uL of the concentrated cells were stained with CD4-V450, CD45-V500, HLA-DR-FITC, CD30-PE, 7AAD, CD19-PE-Cy7, CD14+CD3-APC, CD8-APCH7 and Lymphocyte Screening Tube (Euroflow®). For each sample, 100,000 cells were acquired using FACSCanto-II cytometer and data were analyzed with Infinicyt(tm) software. Positive cases were submitted to a confirmation tube with HLA-DR-V450, CD45-V500, CD45RO-FITC, CD25-PE, CD5-PerCPcy5.5, CD2-PE-Cy7, CD14-APC, CD43-APCH7. Cases with less than 1000 cellular events available in flow cytometry acquisition were considered unavailable. **Results:** 83 PF collection from 77 patients were analysed in 28 months. Five patients had bilateral breast collection and one patient repeated the evaluation 2 weeks after first analysis. Median age was 50 years (31–57 years). We found seven positive cases (9.1% of patients); in one of them, the first sample was considered unavailable. Thus, the MFC sensitivity was 85.7% and specificity 100% in our cohort. From 76 negative samples, 9 (11.8%) were considered unavailable cause of lack of viable cells, 7 (9.2%) were blood contaminated, 11 had neutrophilic exudate (14.5%) and 49 (64.5%) had transudates with a predominance of mature lymphocytes. Cytological examination of all seven positive cases revealed numerous large, anaplastic cells with pleomorphic nuclei, prominent nucleoli, and moderate basophilic cytoplasm with frequent vacuoles. MFC immunophenotyping showed large tumor cells (increased FSC/SSC scatter) with bright expression of CD30, CD25 and HLA-DR, $CD45^{dim}$ and absence of monocytic, B and NK cell antigens (CD14, CD19, CD20, CD38, CD56 and light chain expression). All had absence of CD3, five cases had CD4 heterogeneous expression, one had weak CD8, and one had CD5 dim. In negative cases available, scant or rare CD30 positive lymphocytes with normal morphology was considered reactive and corresponded to activated T cells. Furthermore, when we compared BIA-ALCL and normal cases, we detected a significant MFI difference, with overexpression of CD30, HLA-DR and CD25 and dim expression for T cell markers in tumor cells compared with normal samples. **Conclusion:** Here we describe seven patients with BIA-ALCL and could highlight the utility of cytologic evaluation and multiparametric flow cytometry immunophenotyping in diagnostic workup.

https://doi.org/10.1016/j.htct.2020.10.747

746

DESENVOLVENDO UM ALGORITMO SIMPLES BASEADO EM CITOMETRIA DE FLUXO MULTIPARAMÉTRICA PARA RASTREAMENTO RÁPIDO DE LEUCEMIA PROMIELOCÍTICA AGUDA

V.C. Silva, K.B. Pagnano, G.B. Duarte, M.D.B. Pellegrini, B.K.L. Duarte, K. Metze, I. Lorand-Metze

Centro de Hematologia e Hemoterapia, Universidade Estadual de Campinas (UNICAMP), Campinas, SP, Brasil