

nas amostras analisadas no perfil linfocitário, sendo também possível a aplicação destas em uma maior variedade de casos. Estes métodos computacionais elucidam a identificação de padrões relevantes nas amostras analisadas, o que é especialmente importante em estudos com grandes quantidades de dados, onde torna-se possível a aplicação de estudos de *big-data* e *machine learning*, além do comparativo com a literatura relacionada, a exemplo do entendimento e definição da ontogenia das populações B, T e NK, frente à diferentes quadros clínicos. Por fim, a visualização dos dados celulares pode ser retratada de forma mais completa, com recursos mais variados que podem reforçar o rigor e acompanhamento analítico.

Conclusão: A integração de bibliotecas R especializadas, como *FlowSOM*, *OpenCyto*, e outras, proporcionou uma análise mais robusta e eficiente dos dados de citometria de fluxo. As abordagens avançadas de clustering permitiram identificar as populações B, T e NK, além de caracterizar subpopulações com maior precisão e eficiência, favorecendo o entendimento da ontogenia das mesmas. Essas técnicas têm o potencial de impulsionar significativamente a pesquisa e clínica em citometria de fluxo, trazendo novos parâmetros de análise e abrindo novas possibilidades para o controle interno e entrega dos resultados, por exemplo, na investigação e monitoramento de processos biológicos complexos, além do auxílio no desenvolvimento e acompanhamento de terapias personalizadas.

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ACUTE MYELOID LEUKEMIA WITH A RARE IMMUNOPHENOTYPE AND COMPLEX GENETIC PROFILE

JFDS Tosi, AR Severino, CM Bertolucci,
LGR Barbosa, M Higashi, ER Mattos,
MRV Ikom-Colturato

Hospital Amaral Carvalho (HAC), Jaú, Brazil

Acute Myeloid Leukemia (AML) presents a wide immunophenotypic variability and defining molecular alterations, used for the revised WHO 2022 classification. The genetic profile is also used for risk stratification according to the European LeukemiaNet (ENL) 2021 and treatment definition. Here we present a case with an unusual immunophenotype and complex genetic alterations. Case report: 42-year-old woman, with increased menstrual flow in the last 10 days and no other alterations. On physical examination, she was in good condition, with slight paleness, without hemorrhagic suffusions, absence of lymph node enlargement and hepatosplenomegaly. Complementary tests showed Hb 9.4 g/dL WBC 2850/mm³ (2% blast cells) Platelets 48,000/mm³ Fibrinogen 56 mg/dL (normal: 200 – 400), normal biochemistry and coagulogram. The myelogram showed massive infiltration by hyper granular blasts suggesting basophilic leukemia. Immunophenotyping revealed 3 blast populations, all expressing cyMPObright, CD13, CD33bright, CD34, CD38dim/negative, CD44, CD45intermediate, CD45RA, CD96, CD97, CD99, CD123, CD244, CLL1, TIM3. The differences between the expressions

of the blast populations were: 1st population (41%) expressed partial CD7, CD117bright and homogeneous, heterogeneous HLA-DR (positive to negative), not expressing CD11b and CD203c; 2nd population (40%) expressed CD11b, CD203c bright, without expressions of CD7 and HLA-DR (basophilic component); 3rd population (19%) did not express CD7, CD11b, CD203c and HLA-DR (promyelocytic component). The 3 populations did not express cyCD3, SmCD3, CD4, CD10, CD14, CD15, CD16, CD19, CD36, CD42a+CD61, CD54, CD56, CD64, CD71, cyCD79a, CD105, CD300e. This profile was consistent with AML with basophilic and acute promyelocytic (APL) components. The karyotype was complex: 46,XX, del(3)(p21), del(5)(q15), t(15;17)(q22;q21)[6]/46,XX[14]. The NGS panel revealed the presence of the PML::RARA gene fusion, negative for NPM1, FLT3 ITD and TKD mutations and others. The patient was treated with Cytarabine plus Daunorubicin (7+3) and ATRA. Assessment of measurable residual disease (MRD) by flow cytometry (FC) on D28 of induction showed 0.2% non-APL myeloblasts and 0.29% basophilic blasts. Despite preventive therapies, the patient experienced life-threatening bleeding and sepsis, but had a favorable outcome. She received 2 cycles of consolidation and maintenance therapy. FC MRD and PML::RARA RT-PCR was undetectable after the 2 consolidation cycles and during maintenance therapy. PML::RARA was also undetectable at the end of therapy and 3 months later. The probability of severe bleeding in APL with the concomitant presence of basophilic blasts has already been described, as was seen in this case. The patient had a good therapeutic response. Monitoring MRD in AML is essential to guide treatment, and the methods must be complementary, especially in cases with multiple leukemic clones. The identification of different leukemia clones by immunophenotype reflected the genetic complexity of this case. Therefore, it is important to integrate diagnostic methods for better patient management.

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FLOW CYTOMETRY IMMUNOPHENOTYPING OF HEALTHY PLATELETS AND HOSPITALIZED PATIENTS WITH SUSPECTED PLATELET DYSFUNCTION: CHALLENGES FOR ESTABLISHING A CUTOFF VALUE

DK Cecconello^a, F Spagnol^b, AP Alegretti^b,
DA Pilger^a, MG Farias^b

^a Universidade Federal do Rio Grande do Sul (UFRGS), Porto Alegre, Brazil

^b Hospital de Clínicas de Porto Alegre (HCPA), Porto Alegre, Brazil

Introduction/Objective: Platelets play an essential role in hemostasis, for their characterization, several platelet surface glycoproteins (GP) were identified as GPIIb, evaluated by the expression of CD41, and non-covalently binds to GPIIIa, recognized by the CD61 marker, making up the GPIIb-GPIIIa complex (CD41/CD61). GPIX is identified by CD42a and GPIb by CD42b that interacts together with GPV (CD42d), forming the