and monosomy 7. **Conclusion:** The occurrence of immunophenotypic shifts in B-ALL Ph+ highlights the need for further studies to establish the prognostic value of molecular classifications in these leukemias. This, in turn, will help define the most appropriate therapeutic approach for each subtype.

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NEOPLASIA MIELOIDE COM REARRANJO NO GENE FLT3 REFRATÁRIA QUE APRESENTA COMPROMETIMENTO COM A SÉRIE GRANULOCÍTICA E ERITROIDE

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Paciente do gênero masculino, 20 anos de idade procurou atendimento com quadro de fadiga, tosse produtiva, astenia intensa de padrão progressivo e linfonodomegalias em região cervical bilateral. No hemograma foi evidenciado anemia (4,3 g/dL), leucocitose (52.820/uL) com presença de 84% de blastos e plaquetopenia (17.000/uL). A imunofenotipagem por citometria de fluxo da amostra de medula óssea detectou 51,8% de blastos (CD117+, HLA-DR+FR, CD45+) comprometidos (CD38+) com a linhagem mieloide/granulocítica (MPO+, CD13+, CD33+ +, CD64+FR); além de 2,0% de blastos com comprometimento à série eritroide (CD117+, CD36++, CD105++, CD71++; CD38, HLA-DR, CD42b, CD203c, CD13 e CD33 negativos) e 3,1% de basófilos (CD123+, CD203c+, HLA-DR negativo) com ausência de expressão de CD22. Além disso, foi detectada a presença da mutação FLT3-DIT pelo PCR qualitativo. O paciente realizou o tratamento quimioterápico de indução 7+3. Após 17 dias do início do tratamento, apresentou leucopenia (860/uL) com 6% de blastos em sangue periférico. A análise imunofenotípica da medula óssea evidenciou a presença de 49% de blastos (CD117 +, CD45+, CD34 negativo) comprometidos (CD38+) com a linhagem mieloide/granulocítica (MPO+, HLA-DR+, CD13+, CD33++; CD36, CD71, CD7, CD19 e CD56 negativos) e 9,9% de blastos (CD117+, CD45+, CD34 negativo) comprometidos com a série eritroide (CD36++, CD105++, CD71++; CD38, HLA-DR, CD42b, CD203c, CD13 e CD33 negativos) semelhantes ao diagnóstico. Além disso, foram observadas 2,6% de células da série basofílica (CD203c+, CD33+, HLA-DR negativo), com ausência de expressão de CD22. Do total de basófilos, 50% apresentam características de imaturidade (CD45+FR, CD117+FR, CD13+FR, CD11b+FR). Paciente evoluiu com quadro de neutropenia febril e sepse de provável foco cutâneo (lesões de acne e abscesso perianal) com hemocultura positiva para *Klebsiella pneumoniae* co-produtora de KPC e NDM. Assim, para atingir estabilidade clínica, iniciou protocolo com alta dose de ARA-C associado a inibidor de FLT3. O paciente foi encaminhado ao transplante de medula óssea. A neoplasia com rearranjo no gene FLT3 (fms-like tyrosine kinase 3) é uma doença de células-tronco hematopoiéticas que pode se manifestar com características de SMD, neoplasias mieloproliferativas, mielodisplásicas/mieloproliferativas ou leucemia aguda de linhagem mieloide, linfoide B ou T. Associa-se a um prognóstico desfavorável, com evolução rápida e com alta mortalidade. Os pacientes geralmente apresentam anormalidades em sangue periférico (leucocitose, anemia e plaquetopenia) e podem apresentar linfonodomegalias e esplenomegalia. A maioria dos pacientes é diagnosticado na fase crônica da doença e, os casos em que há mais de 20% de blastos no momento do diagnóstico, devem ser classificados como neoplasia mieloide ou linfoide com a mutação FLT3 em fase blástica. Considerando a necessidade de esclarecimento da linhagem celular afetada, a imunofenotipagem é essencial para um diagnóstico preciso e direcionamento do tratamento. No caso do paciente relatado, as células apresentam comprometimento mieloide/granulocítico/eritroide). O FLT3 é um receptor de tirosina quinase que desempenha um papel importante na proliferação, diferenciação e sobrevida celular e, quando mutado aumenta a proliferação celular e inibe a apoptose. Dessa forma, alguns pacientes obtêm respostas hematológica e citogenética com a utilização de fármacos inibidores de FLT3, no entanto, a resposta parece ser de curta duração e o transplante alogênico é indicado.

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T-CELL CLONES OF UNCERTAIN SIGNIFICANCE/ T-CUS

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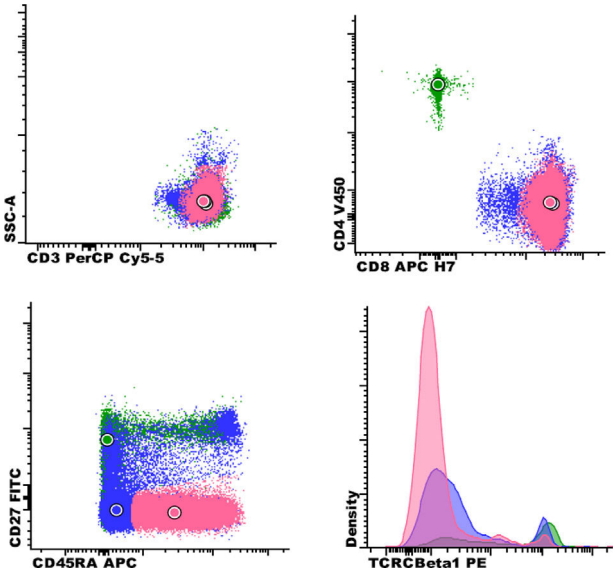
Female patient, 56 years old, with leukocytosis and lymphocytosis and Lymphoma suspicion - Peripheral blood sample with white blood count 23,200 cells / TCD8+ restriction (TCD4/ TCD8 ratio 0.1:1). Panel, Immunophenotyping and Gating Strategy. Triage of all the samples was analyzed in the LST tube. The TCRCBeta 1 tube was designed based on EuroFlow's Orientation Tube for Immunodeficiencies (PIDOT), using CD3, CD4, CD8 and CD45 as "backbones", CD27 and CD45RA to discriminate T-cell subsets, CD16 and CD56 to exclude NK cells, and including TCRCBeta1 in the PE channel. **Results:** 59% T Cells: *52.5% TCD8+: 39.4% TD (terminally differentiated) phenotype with increased absolute levels (9,141 cells/μL - normal age range 8-500) CD7dim CD27(-) CD45RA-/+ TCRCBeta1+5% (monoclonal); 0.7% *Naive* CD45RA+ CD27+ TCRCBeta1+30% (polyclonal), 1.2% CM/TM (central/terminal memory) CD45RA (-) CD27+ TCRCBeta1+19% (polyclonal); 9.7% EM (effector memory) phenotype with increased absolute levels (2,250

cells/μL - normal age range 2-323) CD45RA(-) CD27+ TCRCBeta1+5% (monoclonal). *4.9% TCD4+ TCRCBeta1+44% (polyclonal). **Immunophenotype:** CD45++ CD3+ CD8+ CD38dim CD45RA-/+ CD45RO-/+ CD2+ CD5++ CD7+FR TCR ALPHA-BETA+ PERFORIN+ TCRCBETA1 ++5%; Negative expression: CD25 CD26 CD27 CD28 TCL1 CCR7 TCR GAMMA-DELTA CD11C CD30 CD16 CD56 CD94. **Morphology:** In the analyzed smear, 32% of atypical medium-sized lymphoid cells were observed, with a globose nucleus, generally eccentric, with poorly condensed chromatin with an outline of a nucleolus, and a moderately basophilic, polarized and granular cytoplasm. The inicial diagnostic hypothesis was T-Lymphoma, NOS (the authors judged that there were no sufficient NK antigens expressed to characterize T-LGL). However, further investigation showed the patient had no identifiable T Cell lymphoma, and she eventually was diagnosed with Thrombotic Thrombocytopenic Purpura, with classic symptoms and laboratory confirmation. Since the implementation of TRBC1 for detection of abnormal T-cells, persistent clonal expansion of large granular lymphocytes (T-cell clones of uncertain significance/ T-CUS) have been reported in a variety of clinical conditions, such as hematological neoplasms and autoimmunity. The incidence of T-CUS increases with age, and probably represents reactive T-cell small clones (so called immunoclones), with less than 20% of total lymphocytes or 400 cells/mm3, highly prevalent in patients without T-cell malignancy. Most reactive T-CUS cases are CD8+ or CD4+/CD8+ double-positive.



T-CUS in pink, normal TCD4+ cells in green and normal TCD8+ cells in dark blue.

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POST-TRANSPLANT DOUBLE-POSITIVE T-CELL LYMPHOMA

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Male patient, 54 years old, post-transplant immune profile, bone marrow transplantation for B-cell Non-Hodgkin's-Lymphoma in 2019 - Peripheral blood sample with white blood count 1,800 cells post-transplant immune profile - marked (42%) Double-Positive proliferation. **Panel design, Immunophenotyping and Gating Strategy:** Triage of all the samples was analyzed in the LST tube. The TCRCBeta 1 tube was designed based on EuroFlow's Orientation Tube for Immunodeficiencies (PIDOT), using CD3, CD4, CD8 and CD45 as "backbones", CD27 and CD45RA to discriminate T-cell subsets, CD16 and CD56 to exclude NK cells, and including TCRCBeta1 in the PE channel. **Results:** 57% T Cells: *42% Double-positive CD3+ CD7(-) CD27(-) CD45RA+ TCRCBeta1+100% (monoclonal); *7.5% TCD4+ EM CD45RA(-) CD27(-) TCRCBeta1+ 38% (polyclonal); *7.5% TCD8+ EM CD45RA(-) CD27(-) TCRCBeta1+ 38% (polyclonal). **Immunophenotype:** CD45+ CD3+ CD4+ CD8+ CD26-/+ CD45RA+ CD2+ CD5+ CD11C+ CD57-/+ TCD ALPHA-BETA+ TCRCBETA1+. Negative expression: CD7, CD28, CD45RO, CD25, CD30, TCRGAMA/DELTA, CCR7, CD56, CD16, TCL1. The abnormal Double-positive T-cell population was subsequently detected with a bone marrow sample. The diagnostic conclusion was T-cell Lymphoma, NOS.

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CASE REPORT: BICLONAL SÈZARY SYNDROME

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Introduction: Sézary Syndrome (SS) is defined by the triad of erythroderma, generalized lymphadenopathy, and the presence of clonal T cells with cerebriform nuclei (Sézary cells) in peripheral blood. In addition, one or more of the following criteria are required: an absolute Sézary cell count $\geq$ 1000/μL, an expanded CD4+ T-cell population resulting in a CD4:CD8 ratio of $\geq$ 10, and loss of one or more T cell antigens. T cells have a CD3+, CD4+, CD8(-) phenotype, and characteristically lack CD7 and CD26. The normal counterparts of Sézary cells are circulating central memory T cells (CD27+, CD45RA(-), CD45RO+). According to the degree of circulating involvement, Staging B ("blood") for Sézary syndrome follow these