Introduction/Objective: The diagnosis of mature T-cell lymphoid neoplasms can be particularly challenging due to their overlapping features with reactive T cells. However, the development of specific antibodies targeting the mutually exclusive β-chain isoforms of the T-cell receptor, TRBC1 and TRBC2, has made it possible to assess the restriction of these chains and establish clonality evaluation. This report presents an uncommon case with an immunophenotype suggestive of "T-cell Large Granular Lymphocytic Leukemia," characterized by the presence of two clones: one in the CD4 compartment exhibiting TRBC1 monoclonality and another in the CD8 compartment exhibiting TRBC2 monoclonality. **Case Report:** A 66-year-old male patient with a normal physical examination. The complete blood count showed: hemoglobin 5.27 g/dL, platelets 207,000/mm^3, leukocytes 16,000/mm^3, neutrophils 6,864/mm^3, eosinophils 448/mm^3, monocytes 928/mm^3, and 7,696/mm^3 lymphoid cells with a mature appearance, nuclear irregularities, and abundant granular cytoplasm. A peripheral blood sample was submitted for immunophenotypic evaluation by flow cytometry, which revealed 33.8% T lymphocytes expressing CD2, CD3, CD5 (dim expression), CD56, CD57, and TCR alpha/beta. Of these cells, 20.9% exclusively expressed CD4 and TRBC1, while 12.9% exclusively expressed CD8 and TRBC2, identifying two distinct populations of monoclonal T lymphoid cells with an immunophenotype consistent with T-cell "Large Granular" lymphoid cells. **Discussion:** The diagnosis of mature T-cell lymphoid malignancies is complex and often challenging due to the immunophenotypic overlap between neoplastic T cells and non-neoplastic reactive T cells. The development and implementation of specific antibodies have led to significant advances in clonality assessment. Among these markers, TRBC1 and TRBC2 are especially useful in identifying specific neoplastic clones. The use of these markers has greatly improved the differentiation between neoplastic and reactive T cells, thereby increasing diagnostic accuracy. **Conclusion:** We report a rare case of two distinct mature T-cell lymphoid clones in a case suggestive of "T-cell Large Granular Lymphocytic Leukemia," with few prior cases documented in the literature. This highlights the importance of assessing T-cell clonality using TRBC1 and TRBC2 markers. Advances in this field have been crucial for achieving faster and more accurate diagnoses, which in turn facilitate more effective treatments and better clinical outcomes.

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3

CASE REPORT: B-CELL ACUTE LYMPHOBLASTIC LEUKEMIA (PH+) WITH IMMUNOPHENOTYPIC SHIFT TO MIXED-PHENOTYPE ACUTE LEUKAEMIA B/MYELOID

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Introduction/Objective: The shift in immunophenotype from B-cell Acute Lymphoblastic Leukemia (B-ALL) to Acute Myeloid Leukemia (AML) is a rare phenomenon known as a "phenotypic switch." The aim of this report is to describe a case of B-cell Acute Lymphoblastic Leukemia with an immunophenotypic shift to a Mixed-phenotype Acute Leukaemia B/myeloid. **Case Report:** A 37-year-old male patient presented with complaints of lesions on his tongue, right inguinal region, and buttocks, along with red spots on the skin. One day prior to seeking medical attention, he experienced gum bleeding lasting about an hour. The patient was obese (200 kg). Complete blood count: Hb 10.6 g/dL; Leukocytes 120,290/mm^3 (80% small-sized cells, high nucleus-to-cytoplasm ratio, loose chromatin, evident nucleoli, and agranular basophilic cytoplasm); Platelets 5,000/mm^3. Bone marrow aspiration was not performed due to technical difficulties. Peripheral blood immunophenotyping: Consistent with B-ALL, showing 65.9% low-complexity cells, dim/negative expression of CD45, and positivity for CD19/CD13/CD25/CD33/CD34/CD38/CD58/cyCD79a/CD123/CRFL-2(dim)/TdTnu, with absence of CD3sm/CD3cy/CD10/CD15/CD20/CD22/CD117/cyMPO/cyIgM. Cerebrospinal fluid: 1.6% positive for CD19/CD34/CD38/CD45 and negative for CD3/CD10/CD14/CD20/CD56. FISH: BCR::ABL1 rearrangement t(9;22). Karyotype: 46,XY,t(9;22)(q34;q11.2)[1]/45, idem, -7[19]. Lymphoid panel in peripheral blood: Presence of RUNX1 and BCR::ABL1. The patient was treated with Hyper CVAD + dasatinib and chemotherapy. On Day 20 of treatment, a bone marrow and cerebrospinal fluid reassessment was performed. Bone marrow: 78.0% small to moderately sized cells, high nucleus-to-cytoplasm ratio, nucleus with loose chromatin, and evident nucleoli, occasionally convoluted, basophilic cytoplasm with granules. Immunophenotype: Presence of two cell populations: one with 76.9% myeloid blast cells expressing CD4/CD11b/CD13/CD19/CD33/CD34/CD36/CD38/CD45/CD64/CD71/CD117/CD123/HLA-DR/cyMPO, and negative for CD2/smCD3/cyCD3/CD7/CD10/CD14/CD15/CD20/CD22/CD56/CD61/cyCD79a/IREM-2; and another with 8.3% lymphoid blast cells expressing partial CD10/CD13/CD19/CD22/CD34/CD38/CD58/cyCD79a/CD123/cyIgM (partial), and negative for smCD3/cyCD3/CD117/CD15/CD20/CD25/CD33/CD45/cyMPO/CRFL2. Cerebrospinal fluid immunophenotyping: 13.8% positive for CD34/CD38/CD45/CD64/CD117 and negative for CD3/CD10/CD14/CD19.The patient was treated with FLAG-IDA + Venetoclax + Ponatinib, but developed severe neutropenia, multi-drug-resistant Klebsiella pneumoniae infection, septic shock, and died on Day 17 of the chemotherapy cycle. **Discussion:** The shift in the leukemic cell lineage (lymphoid or myeloid) during the disease course is rare, and the mechanisms involved are not fully understood. These shifts may represent the expansion of a pre-existing clone prior to therapy, clonal evolution, or the development of a new clone. According to the International Consensus Classification of Acute Leukemias 2022, B-ALL Ph+ can be subdivided into two subtypes: "multilineage involvement" and "lymphoid-only involvement." However, there is still no consensus on how to classify these cases. In a large cohort of patients with B-ALL Ph+, Bastian et al. characterized two groups based on transcriptomic and genomic profiles. The "multilineage involvement" group exhibited a genomic pattern of HBS1L deletion

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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4

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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5

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