referenciados ao Serviço de Hematologia de adultos, por diferentes razões, o tratamento transfusional foi suspenso e iniciado hidroxiureia. A mediana (AIQ) de sobrevida após esta mudança de conduta é de 19,5 (15,8-23,3) anos. Vale ressaltar, que dois pacientes perderam o acompanhamento neste período e nestes casos foi considerado apenas o tempo de seguimento. O AVC é manifestação frequente em crianças com doença falciforme, mais especificamente na anemia falciforme. Por vezes, trata-se de uma manifestação catastrófica resultando em sequelas motoras e/ ou neurológicas e impedindo que este indivíduo tenha uma vida normal. Sabe-se que a transfusão de hemácias é tratamento importante para evitar o primeiro episódio (profilaxia primária) e para evitar recidivas (profilaxia secundária). Em nosso relato, as causas de suspensão do esquema transfusional teve várias razões, sendo as mais frequentes: recusa do paciente, dificuldade de comparecer com frequência ao serviço e irregularidade do acompanhamento. Nos casos apresentados, estes pacientes encontram-se há cerca de 20 anos em acompanhamento sem transfusão. O estudo STOP 2 (Optimizing Primary Stroke Prevention in Sickle Cell Anemia Trial) não foi capaz de determinar se a suspensão de transfusão regular pode ser indicada nestes pacientes, e não existe até o momento, nenhum estudo que comprove que a suspensão do regime transfusional e o início de hidroxiureia tenha a mesma resposta na profilaxia. **Conclusão:** Este relato chama a atenção para o fato de haver necessidade de mais estudos controlados para determinar o período mínimo de tratamento transfusional destes pacientes, levando-se em conta as complicações associadas a transfusões frequentes por muitos anos.

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ID – 1922

POTENTIAL IMMUNOMODULATORY FUNCTIONS OF THE ACKR1 GENE IN THE CONTEXT OF AUTOIMMUNE ANEMIAS

FLO Gomes [a,b], JSV Campelo [a,b], CCMX Albuquerque [a,b], LNM Passos [a,b], MR Nascimento [b,c], ACS Castro [d], AN Fujimoto [b], EMC Silva [b], MOO Nascimento [c,e], JPM Neto [a,c,e,f]

[a] *Programa de Pós-graduação em Ciências Aplicadas à Hematologia (PPGH), Universidade do Estado do Amazonas (UEA), Manuas, AM, Brazil*
[b] *Fundação Hospitalar de Hematologia e Hemoterapia do Amazonas (HEMOAM), Manaus, AM, Brazil*
[c] *Programa de Pós-Graduação em Imunologia Básica e Aplicada (PPGIBA), Universidade Federal do Amazonas (UFAM), Manaus, AM, Brazil*
[d] *Programa de Pós-Graduação em Imunologia Básica e Aplicada (PPGIBA), Universidade Federal do Amazonas (UFAM), Manaus, AM, Brazil*
[e] *Programa de Pós-Graduação em Ciências Farmacêuticas (PPGCF), Universidade Federal do Amazonas (UFAM), Manaus, AM, Brazil*
[f] *Universidade Federal de Juiz de Fora (UFJF), Juiz de Fora, MG, Brazil*

Introduction: The Atypical Chemokine Receptor 1 (ACKR1) gene encodes a transmembrane protein found on red blood cells and endothelial cells. This protein functions as a receptor for inflammatory chemokines; however, it is signaling mechanism differs from that of a conventional receptor. Associated with the Duffy blood group system, it also plays a role in inflammation regulation. The FY*BES allele, prevalent among individuals of African descent, results in the cessation of Duffy antigen production, leading to the Fy(a−b−) phenotype, which has been associated with immune system alterations. In autoimmune hemolytic anemia, the presence or absence of the Duffy antigen influences the severity of red blood cell destruction and the subsequent immune response. **Objectives:** The objective of this study is to ascertain the potential role of the ACKR1 gene in the regulation of the immune system and its association with the process of hemolysis, defined as the breakdown of red blood cells in patients diagnosed with autoimmune anemias. **Material and methods:** A comprehensive review of studies from 2019 to 2025 was conducted using the databases PubMed, SciELO and LILACS. The following words were used to describe the project: The keywords “ACKR1”, “chemokines” and “autoimmune hemolytic anemia”, were entered into the search engine. A total of 17 articles were identified using the specified descriptors, however, only five of these were ultimately selected for further analysis. **Discussion and conclusion: Results:** Research demonstrated a correlation between a specific trait, designated as the Fy(a−b−) and phenotype, and the absence of a particular protein on red blood cells, known as Duffy. This absence has significant consequences for the immune system. The manner in which certain cells within the body adhere to one another may undergo alterations, potentially impeding the body’s ability to regulate inflammation and compromising the functionality of the immune system. A substantial body of research has indicated that an insufficient intake of a specific protein, known as Duffy antigen, may result in elevated levels of chemical mediators that trigger inflammatory responses within the body. These elevated levels have the potential to render the immune system more susceptible to self-attack. When combined with genetic susceptibility, this condition can contribute to the breakdown of immune tolerance and the onset of red blood cell destruction caused by autoantibodies. Discussion: This phenomenon can result in an immune system that is more active, which can increase the likelihood of sensitivity to certain substances, particularly in individuals who are genetically predisposed to such reactions. Conclusion: The absence of Duffy antigen expression on red blood cells may play a significant role in the process of inflammation and the autoimmune causes of certain types of anemia. Genetic testing for the Duffy gene in patients with AIHA enhances medical professionals’

understanding of the condition and improves knowledge of potential autoimmune disease risks.

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PREDICTING VASO-OCCLUSIVE CRISIS RISK IN SICKLE CELL DISEASE BY MEASURING PROTECTION, NOT JUST HBF: SINGLE-CELL QUANTIfiCATION OF FETAL HEMOGLOBIN WITH AN OPTIMIZED SPIKE-IN FLOW CYTOMETRY ASSAY

ML Arrojo [a], GBC Moreto [a], J Milhomens [b], CCOM Bonaldo [b], ACS Pinto [b], SK Haddad [b], RA Panepucci [a]

[a] *Laboratory of Functional Biology (LFBio), Ribeirão Preto, SP, Brazil*
[b] *Regional Blood Center, Ribeirão Preto, SP, Brazil*

Introduction: Accurate quantification of fetal hemoglobin (HbF) at the single-cell level is essential for evaluating functional protection of red blood cells (RBCs) in sickle cell disease (SCD) and for monitoring therapeutic interventions. Conventional metrics—%HbF by HPLC and F-cell analysis—lack the resolution to directly estimate mean cellular HbF (cHbF) and precisely identify functionally protected RBCs (%ProtRBC; cHbF > 10 pg), a parameter strongly associated with reduced sickling under hypoxia. **Objectives:** To develop and validate a novel flow cytometry-based assay for quantifying fetal hemoglobin content (cHbF) at the single-cell level, and to demonstrate its clinical applicability for predicting vaso-occlusive crises (VOC) in patients with sickle cell disease. **Material and methods:** RBCs from 17 HbSS patients were analyzed by quantitative flow cytometry using an optimized Spike-In assay. Spike-In calibrators were prepared by mixing normal RBCs with RBCs from a hereditary persistence of HbF (HPFH) SCD donor, generating two reference populations. In each assay, 9×10^5 patient RBCs were mixed with 1×10^5 fluorescently labeled Spike-In RBCs, fixed, stained with anti-HbF antibodies, acquired on a BD FACSymphony A1, and analyzed in FlowJo. Internal calibration curves converted anti-HbF fluorescence into cHbF values at the single-cell level. %ProtRBC was compared to %HbF by HPLC and classical F-cell counts. Predictive performance for vaso-occlusive crises (VOC) was evaluated by ROC analysis. Agreement between Spike-In–derived MCHbF and values calculated from HPLC-derived %HbF × MCH was assessed by correlation, regression, and Bland–Altman analysis. Assay robustness was tested with two different HPFH Spike-In donors. **Results:** Eight patients experienced a VOC within six months. The optimized Spike-In assay yielded the highest discriminative capacity for VOC prediction (AUROC = 0.958), with 87.5% sensitivity (95% CI: 52.9–99.3%), 88.9% specificity (95% CI: 56.5–99.4%), and $LR^+ = 7.9$ at %ProtRBC < 27.35, outperforming classical F-cell (AUROC = 0.875; $LR^+ = 3.9$) and %HbF by HPLC (AUROC = 0.819; $LR^+ = 2.6$). At cut-offs achieving 100% sensitivity, Spike-In retained the highest specificity (66.7%; $LR^+ = 3.0$) versus F-cell (44.4%; $LR^+ = 1.8$) and HPLC (11.1%; $LR^+ = 1.1$). Spike-In cHbF strongly correlated with MCHbF from HPLC × MCH ($r = 0.7401$; $p = 0.0002$), with proportional agreement when regression was constrained through zero (slope = 0.8477; 95% CI: 0.7085 –0.9869). Bland–Altman analysis indicated minimal bias (–0.84 pg) and narrow limits of agreement (–7.81 to 6.12 pg), confirming accuracy across the measurement range. cHbF values obtained with different HPFH Spike-In donors showed almost perfect correlation ($r = 0.9828$; $p < 0.0001$), demonstrating high reproducibility and portability of the method. **Discussion and conclusion:** The optimized Spike-In flow cytometry assay provides accurate, high-throughput, single-cell quantification of cHbF and superior prediction of VOC risk compared to standard methods. Its strong agreement with reference calculations, reproducibility across calibrators, and ability to resolve functional RBC heterogeneity establish it as a robust, clinically relevant biomarker platform for SCD management and for assessing pharmacological or genetic HbF-inducing therapies. This study was financed, in part, by the São Paulo Research Foundation (FAPESP), Brazil (Process Number: #2022/12856-6).

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PREVALENCE AND IMPACT OF ANEMIA DUE TO HEMOLYSIS IN HIGH-PERFORMANCE ATHLETES

GM Farias [a], RS Leal [b], ÉJS Freitas [c,d], MO Cunha [d], AGS Gbadamassi [c,d], VB Lima Lobato [d], MR Nascimento [e], NA Fraiji [e], R Ramasawmy [c], JPM Neto [c,d,e]

[a] *Universidade Federal da Bahia (UFBA), Salvador, BA, Brazil*
[b] *Pós-Graduação em Farmácia (PPGFAR), Universidade Federal da Bahia (UFBA), Salvador, BA, Brazil*
[c] *Programa de Pós-Graduação em Imunologia Básica e Aplicada (PPGIBA), Universidade Federal do Amazonas (UFAM), Manaus, AM, Brazil*
[d] *Programa de Pós-Graduação em Ciências Farmacêuticas (PPGCF), Universidade Federal do Amazonas (UFAM), Manaus, AM, Brazil*
[e] *Universidade Federal de Juiz de Fora (UFJF), Juiz de Fora, MG, Brazil*

Introduction: Engaging in moderate to intense physical activity has been demonstrated to trigger hematological alterations, among which anemia is one of the most common and clinically relevant conditions. This condition is marked by a decrease in hemoglobin concentration within blood, leading to impairing oxygen transport and consequently diminished physical performance. Anemia in athletes, can be attributed to a variety of factors, including exercise- induced hemolysis, gastrointestinal or urinary losses, iron deficiency, and nutritional inadequacies. **Objectives:** This study aimed to review the scientific literature concerning the mechanisms,