ELSEVIER

Blood 142 (2023) 2454

The 65th ASH Annual Meeting Abstracts

POSTER ABSTRACTS

101.RED CELLS AND ERYTHROPOIESIS, EXCLUDING IRON

Erythrophagocytosis Mediated By Circulating Monocytes Is Induced By Distinct Mechanisms Depending on the Disease

Marina Dorigatti Borges, PhD[1], *Dulcineia Albuquerque, PhD*[1], *Carolina Lanaro, BSc, PhD*[1], *Sara Teresinha Olalla Saad, MD*[1], *Katia B Pagnano, MDPhD*[1], *Renata Sesti-Costa, PhD*[1,1], *Fernando Ferreira Costa, MD PhD*[2]

[1] Hematology and Hemotherapy Center, University of Campinas - UNICAMP, Campinas, Brazil
[2] Hematology and Transfusion Center, University of Campinas - UNICAMP, Campinas, Brazil

Stress erythropoiesis (SE) occurs when the steady state erythropoiesis is no longer sufficient to sustain the number of healthy red blood cells (RBC) needed in the circulation. There is an extension of this activity to extramedullary sites, contributing to the increase in cell production in the erythroblastic island (EBI) niche, and altered or damaged RBCs are then cleared by macrophages. Some diseases have a state of chronic SE. Sickle cell anemia (SCA) affects multiple aspects of RBCs, leading to recurrent vaso-occlusive crisis, hemolytic anemia, and inflammation. Polycythemia Vera (PV), an acquired neoplastic disease with mutations in JAK2 kinase, is characterized by the overproduction of RBCs, increasing blood viscosity, and resulting in a higher risk of thrombotic events. We previously showed that circulating monocytes (MC) of both diseases have a phenotype similar to the EBIs macrophages, with higher expression of CD169, CD163, CD206 and VCAM-1, and also participate in the phagocytosis of RBCs, function typically carried out by macrophages. Therefore, the aim of this study was to determine whether the increased phagocytic activity found in these patients was due to the alterations on the MCs phenotype or if it was an influence of the RBCs, as well as investigate the consequences to the iron metabolism after phagocytosis. MCs from peripheral blood were isolated and incubated for 2 hours with previously stained RBCs, to allow the phagocytosis to happen. After this period, the remaining RBC were lysed and the antibodies anti-intracellular heme-oxigenase-1 (HO-1) and anti-membrane ferroportin (FPN) were used for flow cytometry analysis. The statistical analysis was done by paired t-student test ($P<0.05$). When exposed to SCA RBCs, MCs from SCA patients and healthy controls (HC) show higher phagocytic capability, than when exposed to HC RBCs (MC_{HC} 22.9±8 vs 9.4±1.1, **P=0,0099; MC_{SC}A 33.6±11.4 vs 16.4±11, **P=0,0054; respectively). The same happens to PV and HC MCs: when exposed to PV RBCs: they show higher phagocytosis compared to the presence of HC RBCs (MC_{HC} 9.3±4.4 vs 2.8±4.7, **P=0.0065; MC_{PV} 21.3±6.1 vs 11±6.8, *P=0.0146; respectively). Investigating the role of MCs in this activity, we compared SCA, PV and HC MCs in the presence of patients RBCs. SCA MCs internalized RBCs similarly to HC MCs (P=0.0699); on the other hand, PV MCs presented higher phagocytic capability when compared to HC MCs (**P=0.0022). In all groups, the expressions of both HO-1 and FPN were higher in those MCs that participated in the phagocytosis than in those that didn't contribute to this function (HO-1: MC_{SCA} 91.9±4 vs 65.9±16.8, **P=0.0098, MC_{PV} 92.1±5.5 vs 61.7±15.5, ***P=0.0007; FPN: MC_{SCA} 81.8±13.8 vs 66.5±15.7, ***P=0.0003, MC_{PV} 70.9±17 vs 53.2±16.5, **P=0.0013 respectively). This indicates an increase in the ability to degrade heme and export iron post-phagocytosis. The results suggest that RBCs from patients with SCA are the ones responsible for the increase in the phagocytic activity by MCs, indicating that RBCs clearance is not an innate function of SCA MCs, even though they are active participants in the circulation. For PV, both RBCs and MCs seem to contribute to the increase of this process in the circulation of patients. The higher expression of molecules related to the iron metabolism suggests a participation of circulating MCs to the iron disponibility, helping with the maintenance of stock for the erythrocytic production. Even though SCA and PV have different pathophysiology, they both present SE and the circulating MCs from both groups show an important participation in the clearance of RBCs, either by an intrinsic function of MCs or a direct influence of changes in the RBCs, suggesting the important contribution of MCs to avoid oxidative stress and help with iron disponibility for erythropoiesis during the course of SE.

Disclosures Saad: *FAPESP Sao Paulo state foundation:* Research Funding. **Costa:** *Novartis Pharma AG:* Honoraria; *Pfizer:* Consultancy.

https://doi.org/10.1182/blood-2023-189370