Distinct plasticity of alveolar macrophages during pulmonary malaria: establishing new targets for future therapeutic approaches

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**Introduction:** Malaria is an infectious and parasitic disease and one of the main public health challenges, affecting 229 million people worldwide. The parasitosis could progress to severe forms, such as cerebral malaria, severe anemia, and malaria-associated with acute respiratory distress syndrome (M-ARDS). M-ARDS is observed in humans, in experimental murine models, and is characterized by pulmonary microvascular injury leading to increased permeability of endothelium and inflammation. However, there is a lack of knowledge about immunoregulatory mechanisms involved in the pathogenesis development. Thus, the aim of this study was to investigate the recruitment kinetics of innate immune cells into the lung and polarization of alveolar macrophages in murine models of malaria.

**Methods:** To this end, C57BL/6 and BALB/c mice were inoculated with blood containing Plasmodium berghei ANKA (PbA)-infected erythrocytes. At 4 and 6 days post infection (dpi), clinical parameters were evaluated. Then, animals were euthanized, the bronchoalveolar lavage (BAL) was performed and was collected for analysis of inflammatory biomarkers, such as cytokines and nitric oxide. To assess the cells in the BAL and to identify myeloid cells in the lung tissue the cytometry was performed.

 **Results:** C56BL/6 infected mice showed pulmonary dysfunction and edema, evidenced by increased organ weight and BAL protein content when compared to BALB/c infected mice. In addition, we demonstrated that C56BL/6 infected mice developed alterations in plethysmography (pause and respiratory rate). However BALB/c infected mice exhibited a slight increase in organ weight and proteins in the BAL. These alterations were not reflected in lung dysfuction. We observed differences in the percentage and total number of inflammatory monocytes and neutrophils, in BAL and tissue in the two analyzed models. Both infected strain mice showed percentage reduction and total number of alveolar macrophages. Interestingly, we observed that alveolar macrophages from C57BL/6 mice exhibited higher expression of the macrophage marker with M2 profile (CD206) and a reduction in the percentage of cells expressing the enzyme Inos, a macrophage marker M1. Furthermore, we observed a significant increase in the activity of M1 macrophages, arginase enzyme and nitric oxide in the BAL of C57BL/6 infected mice. The analysis of cytokines in the BAL in both strains revealed increased levels of the proinflammatory cytokines TNFα and IFNγ .However, in the BALB/c infected mice we also noticed an increase in the anti-inflammatory cytokine IL-10.

**Conclusion:** Taken together, our data demonstrated that C57BL/6 and BALB/c infected mice have differentiated cell population dynamics in the lung, with distinct plasticity of alveolar macrophages and an imbalance between pro and anti-inflammatory cytokines. Thus, the understanding of this dynamic could suggest the establishment of new targets for future therapeutic approaches to treat the pulmonary malaria, acting mainly in the control of the pulmonary damage, and improving the quality of life of affected patients worldwide.

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