Instituto de Medicina Molecular João Lobo Antunes

Master Project Proposal

Title: Parasite exploitation of host immune complement, C1s, during malaria infection

Synopsis:

Malaria still kills a child every 2 minutes. *Plasmodium falciparum*, the most virulent *Plasmodium* parasite, is responsible for more than 95% of severe illnesses and deaths due to malaria[1]. Cerebral malaria is a frequent manifestation of a severe *P. falciparum* infection in children and is often fatal. A major feature observed in cerebral malaria is the sequestration of *P. falciparum* infected erythrocytes (IEs) in the host microvasculature. Indeed, *P. falciparum* has the unique ability to bind to host endothelial cells to avoid clearance by the spleen; however, this results in obstruction of the microvasculature and inflammation[2]. It is well established that the parasite variant antigen, *P. falciparum* erythrocyte membrane 1 (PfEMP1) expressed on the surface of the IEs, mediates binding of parasites to host microvascular endothelial cells[3]. PfEMP1 is expressed by a family of approximately 60 *var* genes.

Our preliminary data show that human serum-derived complement C1s cleaves the conserved interdomain regions of most PfEMP1 types, resulting in the removal of receptor binding domains and preventing cytoadhesion and sequestration of IEs[4]. C1s is a subunit of complement component 1 in the classical complement pathway. It is a serine protease which cleaves and activates complement C2 and C4 initiating proteolytic activity of the pathway. C1s can also cleave non-complement proteins such as major histocompatibility complex class I antigens and insulin-like growth factor-binding protein[5]. The conservation of the C1s cleavage site in an otherwise highly variant PfEMP1 suggests that *P. falciparum* parasites have been selected to maintain susceptibility to cleavage, possibly in order to modulate the infection towards increasing parasite survival while controlling damage to the host.

We now aim to determine the mechanism by which the parasite exploits the host complement C1s during infection and disease. In the present proposal, the selected candidate will (i) determine how C1s is regulated during malaria infection using clinical samples, (ii) analyze the role of PfEMP1 domain cleavage in immune evasion and (iii) assess the ability of C1s levels to drive the expression of particular *P. falciparum var* genes. The laboratory techniques that will

be used include ELISAs, cell culturing, flow cytometry, RT-PCR, cytoadhesion and phagocytosis assays.

Results arising from this project will potentially have a broad impact as it will unravel a novel function of C1s during infection and a pathway for parasite immune evasion. Elucidating how the parasite uses C1s to ensure its survival will open up therapeutic possibilities targeting IE sequestration, a major pathological feature at the basis of severe disease.

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Bibliography:

1. Geneva: World Health Organization. World Malaria Report 2021. (2021).
2. Dondorp, A. M. *et al.* Direct in vivo assessment of microcirculatory dysfunction in severe falciparum malaria. *J Infect Dis* **197**, 79–84 (2008).
3. Lee, W.-C., Russell, B. & Rénia, L. Sticking for a Cause: The Falciparum Malaria Parasites Cytoadherence Paradigm. *Frontiers in Immunology* **10**, 1–15 (2019).
4. Azasi, Y. *et al.* Complement C1s cleaves PfEMP1 at interdomain conserved sites inhibiting Plasmodium falciparum cytoadherence. *Proceedings of the National Academy of Sciences* **118**, e2104166118 (2021).
5. Gál, P., Ambrus, G. & Závodszky, P. C1s, the protease messenger of C1. Structure, function and physiological significance. *Immunobiology* **205**, 383–94 (2002).