FCT Fundação para a Ciência e a Tecnologia
MINISTÉRIO DA CIÊNCIA, TECNOLOGIA E ENSINO SUPERIOR

PTDC/SAU-MIC/117060/2010

Utilization of host cell resources by the malaria parasite in the liver

Malaria is a devastating disease. Once thought to be close to eradication, it is now responsible for nearly half a billion infections and one million deaths every year. The first stage of mammalian infection by *Plasmodium*, the causative agent of malaria, occurs in the host's liver. Although clinically silent, this phase of infection is obligatory and is responsible for an increase in parasite numbers of at least two orders of magnitude. Its asymptomatic nature makes the liver stage of *Plasmodium* infection an ideal target for prophylactic intervention. However, this is arguably the least studied and understood stage of the parasite's life cycle and one where several important questions remain unanswered.

The host laboratory has been at the forefront of *Plasmodium* liver stage research, having made significant contributions to further the understanding of the interactions that take place at the host-parasite interface. We have unveiled crucial aspects of hepatocyte infection by *Plasmodium*, developed techniques to study the parasite's infection of liver cells and identified key host factors that influence its outcome. In the present project we propose to unveil hitherto unknown aspects of *Plasmodium* liver infection, which can be summarized by the question: how does *Plasmodium* subvert its host to gain access to the essential nutrients it requires for intra-hepatic development, to regulate homeostasis of its host cell, and to ensure survival of the host cell?

We set out to answer this question based on several published observations from our laboratory alongside an array of robust preliminary results. Specifically, we have shown that (i) host membrane transporters are modulated during *Plasmodium* development and intracellular parasites activate volume-regulated anion channel-like activity in the host cell plasma membrane; (ii) the plasmodial glucose transporter is essential for development of *Plasmodium* liver stages and glucose enhances *Plasmodium* intra-hepatic development while leading to increased survival of infected host cells; (iii) a number of nuclear receptor (NR) ligands and synthetic NR agonists or antagonists markedly influence development of *Plasmodium* in human hepatoma cells and RNAi-mediated down-modulation of several NRs impairs infection of liver cells by *Plasmodium*. These observations raised three hypotheses that we now propose to address: (I) *Plasmodium* uses specific host transport proteins to regulate ionic balance, maintain homeostasis and fulfill its nutritional needs; (II) *Plasmodium* modulates uptake of glucose by hepatocytes in order to develop and to ensure host cell survival; and (III) Nuclear receptor signaling pathways regulate homeostasis while ensuring a continuous supply of nutrients to the parasite.

In order to test these hypotheses, we will use our own expertise alongside that of our collaborators and consultants in this project. We will employ state-of-the-art methodologies including immunofluorescence confocal microscopy, RNA interference, quantitative real-time PCR, and patch-clamping. These will be used in combination with established *in vitro* models of infection as well as rodent models of malaria, both of which the host laboratory has a vast experience with. This will enable us to address experimentally key aspects of solute and ion transport across infected cell membranes, energy acquisition by the parasite, control of homeostasis, and regulation of cell survival.

We expect that our studies will reveal fundamental aspects of liver infection by *Plasmodium* and its interaction with its host cell. Furthermore, we expect to identify host molecules and pathways that can be exploited as anti-malarial targets. The potential of identified druggable host factors will be investigated and may lead to novel prophylactic strategies against malaria. This is a particularly timely concern given that the liver stage of infection is an ideal target for prophylactic intervention and one for which there is an obvious shortage of effective compounds.