# OUTLINE OF THIS THESIS

In order to test the hypothesis that *P. falciparum* parasites can modify the permeability of the membrane to specific solutes by epigenetic regulation of *clag3* genes expression, in **article 1** of this thesis we explored the role of switches in *clag3* expression in the acquisition of resistance to the antibiotic BS. To fulfil this aim, we selected 10G parasites, which under standard culture conditions express *clag3.2,* under different concentrations of BS. We observed that drug pressure at low concentrations selected for parasites expressing *clag3.1,* whereas parasites exposed to higher concentrations of BS had repressed the expression of both *clag3* genes. Thus, we concluded that parasites develop resistance to BS through changes in their *clag3* gene expression pattern: those parasites expressing *clag3.1* presented higher values of $IC_{50}$ than parasites expressing *clag3.2*; and those parasites not expressing any *clag3* were almost completely resistant to the drug. We did not find any mutation in the genome of these parasites that could explain the change in their phenotype. Hence, we concluded that this mechanism of drug resistance is regulated at the epigenetic level, being the first one of this kind to be described in *Plasmodium* parasites.

After the results obtained in lab-adapted strains and due to the potential significance of this new drug resistance mechanisms in clinical malaria, in **article 2** we investigated the dynamics of *clag3* genes expression in human infections. We found that parasites collected from patients with uncomplicated malaria predominantly express one of the two paralogues, consistent with the property of mutually exclusive expression previously observed in culture-adapted parasite lines. Interestingly, parasites from all the isolates analyzed were expressing the same gene: *clag3.2*. Then, we adapted two parasite isolates from natural-infections to standard-culture conditions. We also selected them under BS pressure. We detected different patterns of selection depending on the isolate. These results lead to the idea that solute transport efficiency, specific of each *clag3* paralogue, depends on the genetic background of the parasite, and is probably determined by the polymorphic regions of *clag3* sequences.

Our studies with BS revealed a new mechanism of drug resistance in malaria parasites. However, this antibiotic is not used in clinical malaria for its high toxicity to human cells. For this reason, in **article 3** we tested whether other drugs could be susceptible of failure by this drug resistance mechanism. For our experiments we used compounds that are suspected to require facilitated transport to reach the cell. We observed that the uptake of three of these compounds requires, at least partially, *clag3* expression, suggesting that parasites could become resistant to these compounds through epigenetic regulation of *clag3* genes. The rest of the drugs that we tested might use alternative routes in which *clag3* genes are not involved.