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Drug targets in the malaria parasite, *Plasmodium falciparum*

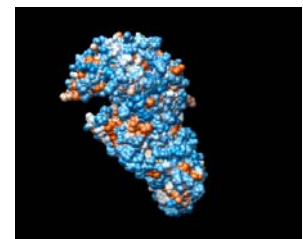
Our laboratory is interested in parasitic diseases, with a primary focus on the causative agent of severe malaria, *Plasmodium falciparum*. The burden of disease-causing parasites is particularly high in developing countries, and inadequate resources are directed towards the development of much-needed treatments. Complete genome sequences are available for many of these parasites, so a wealth of information is available from which to search for potential targets for chemotherapeutic interventions. We are interested in identifying and characterising promising drug targets from *P. falciparum* and other parasites, as well as studying the modes of action and mechanisms of resistance for existing drugs.

We use various bioinformatic methods to screen for promising drug targets, and molecular biological, and cell biological methods (confocal and electron microscopy with transgenic parasites), as well as *in vitro* parasite culture to characterise and test potential parasite drug targets. In collaboration with Professor Malcolm McConville, we are subjecting malaria parasites to metabolomic analyses to help understand the modes of action of existing and novel anti-malarial drugs.

Aminoacyl-tRNA synthetases enzymes as drug targets:

Projects on offer are aimed at characterizing aminoacyl-tRNA synthetase (aaRS) enzymes as drug targets in *Plasmodium*. These enzymes catalyse the attachment of amino acids to their relevant tRNA molecules and are essential for protein synthesis. They have recently been recognized as promising drug targets across a broad range of microbes, and we have recently identified *Plasmodium* aaRSs that are potential targets for new drugs to treat malaria. *Plasmodium* aaRS enzymes differ from those of humans, so we hope to develop drugs specific for *Plasmodium*. We are using *in silico* screening methods to identify likely inhibitors of *Plasmodium* tRNA synthetases and developing assays to measure specific inhibition of *Plasmodium* aaRS enzymes. We will also test inhibitors for their ability to kill *Plasmodium* grown in culture.

Fig. 1. A structural model of a *P. falciparum* aminoacyl-tRNA synthetase. aaRS enzymes have several deep substrate-binding pockets that appear to be suitable for targeting with small molecule inhibitors. Such inhibition has already been demonstrated for many bacteria, including *Staphylococcus aureus* (golden staph).



The metabolic response of *Plasmodium* parasites to antimalarial drugs

Prof. Malcolm McConville and Dr Jim Macrae have developed methods to analyse the metabolome of *P. falciparum* using chromatographic separation of small molecule metabolites and mass spectrometry. The methodology will allow us to sample much of the parasite's metabolome before and after treatment with antimalarial drugs. We will explore the mode of action of some well-known antimalarials, as well as potential targets for promising but uncharacterized novel antimalarials.

P. falciparum-specific virulence factors

The availability of genome sequences for *P. falciparum* and *P. vivax*, another malaria-causing species, allows us to hunt for the molecules underlying the differences in virulence and pathogenesis between these organisms. We have identified a *falciparum*-specific gene; a nucleoside triphosphate diphosphohydrolase

(NTPDase) that we hypothesize encodes a parasite virulence factor that modulates host inflammation and immune responses. In collaboration with Dr Elizabeth Hartland (Dept of Microbiology, University of Melbourne) we will characterize the *P. falciparum* NTPDase by studying its localisation, activity and susceptibility to inhibitors.

Nuclear biology in *P. falciparum*

P. falciparum successfully evades the human immune system by an ingenious trick of constantly changing the surface coat to avoid detection and destruction mediated by human antibodies. This transformation is referred to as antigenic variation, and is controlled by a fascinating mechanism of genetic regulation. Many of the regulatory processes involved in this mechanism are epigenetic, ie dependent on modifications of the nucleosomes that serve as a scaffold for DNA wrapping.

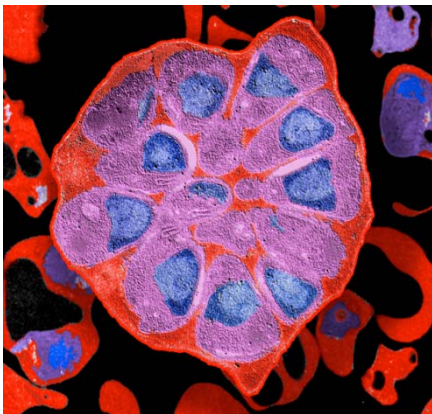


Fig. 2. False-coloured transmission electron micrograph of a *P. falciparum* parasite, cause of the most severe form of malaria. Antigenic variation in these parasites contributes to parasite immune evasion, and compartmentalization of silencing factors and chromatin states in the nucleus play important roles in regulating antigenic variation. In this image, the infected erythrocyte is shown in red, and the parasite is shown in purple. The dark blue highlights the condensed material at the nuclear periphery with lighter blue displaying the nuclear core

Chromosomal compartmentalization in *P. falciparum*

Genetic elements within and adjacent to genes help regulate the covalent modifications of the nucleosomes to which they are attached. These elements also delineate where one nucleosome-modification zone ends and where the next begins. This phenomenon has been very poorly studied in any microbe, but is extremely important for understanding how some genes are highly activated while others are silenced. We have established genetic screens to identify the nucleosome-regulating genetic elements in *P. falciparum*, and will identify the proteins that recognise these elements.

Upstream elements regulating transcription in *Plasmodium*

While much is known about the genetic elements that regulate transcription in some model organisms, transcriptional promoters are poorly understood in *Plasmodium*. Knowledge of the sequence of the *Plasmodium* genome combined with a number of whole-genome microarray experiments enable us to identify the genetic elements that regulate timing and abundance of *Plasmodium* transcription. This project will combine bioinformatic analyses with parasite genetic transfection experiments to characterize transcriptional regulation.

Recent publications

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2. Ralph SA and Scherf A. (2005) 'The epigenetic control of antigenic variation in *Plasmodium falciparum*.' *Curr Opin Microbiol*, 8, 434–440.
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