

P018 Modelling, cloning and expression of the *P. falciparum* NADH dehydrogenase

Nick Fisher, Stephen Ward and Giancarlo Biagini

*Liverpool School of Tropical Medicine,
Liverpool L3 5QA, UK.*

The respiratory chain of the human malaria parasite *Plasmodium falciparum* lacks a canonical protonmotive NADH:ubiquinone oxidoreductase (Complex I), containing instead a single-subunit, non-protonmotive Ndh2, similar to that found in plant mitochondria, fungi and some bacteria. As such, the *P. falciparum* Ndh2 (PfNdh2) presents itself as an attractive anti-malarial chemotherapeutic target, and we have developed a structural model and heterologous expression system for this enzyme to facilitate its physicochemical and enzymological characterisation.

Structural modelling of PfNdh2 suggests the presence of two Rossmann folds forming the flavin and NADH binding sites, with membrane attachment *via* a C-terminal amphipathic helix. Heterologous expression of PfNdh2 in *E.coli* NADH dehydrogenase knockout strain ANN0222 yields an active enzyme with a K_m for NADH of 14 μM in crude membrane preparations. In addition, the recombinant PfNdh2 is strongly inhibited by 1-hydroxy-2-dodecyl-4(1H)quinolone ($\text{IC}_{50} = 50 \text{ nM}$ in crude membrane preparations) with decylubiquinone as substrate. Purification of His-tagged PfNdh2 for high throughput drug screening, and NMR/FTIR spectroscopic investigation is currently underway.