Malarial protein kinases - PfPK7 an atypical mitogen activated protein kinase kinase.

[1] **J.P.Dean Goldring**, [2]Dominique Dorin and [2]Christian Doerig

[1]Biochemistry, University of KwaZulu-Natal, Pietermaritzburg, South Africa; [2]INSERM U609, Wellcome Centre for Molecular Parasitology, Anderson College, 56 Dumbarton Road, Glasgow G11 6NU, Scotland.

Malaria affects 300 million people each year. An effective malaria vaccine has not been developed, immunotherapy appears unlikely (Goldring, 2004) and new therapeutic strategies are needed due to the spread of multi-drug resistant parasites. Malaria parasites undergo many rounds of DNA replication producing multinucleated schizonts. The malaria cell cycle has unusual characteristics which are likely to be due to distinct attributes of the enzymes involved. We have begun to characterise malaria protein kinases involved in cell signalling and regulation of the cell cycle. Malaria kinases are potential therapeutic targets. Using *in silico* screening of the PlasmoDB database potential malarial kinases have been identified. From the deduced amino-acid sequences immunogenic peptides were selected, synthesized and chicken anti-peptide antibodies raised and affinity purified. The anti-peptide antibodies were used to identify and isolate native and recombinant kinases. Kinase activities were determined with specific substrates and inhibitors. Plasmodial mitogen activated protein kinases (MAPK, Doerig et al 1996, Dorin et al 1999) and plasmodial cyclins (Merckx et al 2003) have been characterised. *Plasmodium falciparum* protein kinase 7 (PfPk7) phosphorylates myelin binding protein, histone H2A and beta casein, but not histone H1, alpha casein or malarial MAPK. PfPL7 is expressed during the mosquito and erythrocytic stages of parasite development. The carboxy terminus of the enzyme has homology to MAPKkinases and the amino terminus to fungal protein kinases. PfPK7 reflects the absence of a classical three component MAPK. MAPKK, MAPKKK eukaryotic pathway (Raman and Cobb, 2003) in malarial parasites.

C.M. Doerig, D. Parzy, G. Langsley, P. Horrocks, P. Carter and C.D. Doerig. (1996) A MAP kinase homologue from the human malaria parasite, *Plasmodium falciparum.* Gene, 177, 1-6.

D. Dorin, P. Alano, I. Boccaccio, L. Ciceron, C. Doerig, R. Suplice and D. Parzy. (1999) An atypical mitogen activated protein kinase (MAPK) homologue expressed in gametocytes of the human malaria parasite *Plasmodium falciparum.* Identification of a MAPK signature. Journal of Biological Chemistry 274, 29912 - 20020.

J.P.Dean Goldring (2004) Evaluation of immunotherapy to reverse sequestration in the treatment of severe *Plasmodium falciparum* malaria. Immunology and Cell Biology 82, 447 - 452.

A. Merckx, K. Le Roch, M-P. Nivez, D. Dorin, P. Alano, G. J. Guiterrez, A. R. Nebreda, D. Goldring, C. Whittle, S. Patterson, D. Chakrabarti, and C. Doerig. (2003) Identification and initial characterization of three novel cyclin-related proteins of the human malaria parasite *Plasmodium falciparum.* Journal of Biological Chemistry. 278: 39839-39850.

M. Raman and M.H. Cobb. (2003)MAP kinase modules: many roads home. Current Biology 13, R886 - R888.