A216

Protection against malaria in toque monkeys immunized with P. cynomolgi MSP1inv in alum

Immunization of toque monkeys with baculovirus-expressed, His –tagged recombinant *Plasmodium cynomolgi ceylonensis* (Pcc) C-terminal 19 kDa proteins of the major merozoite surface protein 1 (MSP 1inv) with Freunds adjuvant, mediates long-term protection against homologous and heterologous *P. cynomolgi* blood-stage challenge infection. However, Freunds adjuvant is unsuitable for use in humans, which necessitates the testing of alternative adjuvants. Antigen-alum formulation and binding was optimized for the new preparation of metalloaffinity-purified MSPI inv) antigen under conditions acceptable for human trials, including 800ug aluminium per dose, in accordance with the permitted FDA maximum. This formulation was used in an immunization trial using the P. cynomologi-toque monkey system, which is analogous to the P.vivax-human system. Group 1 comprising 4 animals were immunized with alum+MSP1 inv,

and group 2 comprising 3 monkeys received alum alone. Four doses of immunization were given intramuscularly at 0,1,3 and 4 month intervals. After immunization, the anti-MSP inv antibody titres of immunized animals reached 2.8x104. All animals were given a homologous Pcc challenge infection one month after the last dose of immunization. One of the four immunized animals was completely protected while the other 3 animals showed low patent parasitaemia, resulting in an overall partially protective effect 9p=0.02). immunization with alum did not result in strerile immunity, as seen with Freunds, where antibody titres range from 10^6-10^7 .

Following treatment, the animals were given a second heterologous, blood-stage challenge infection of *P. cynomolgi* Gombak, (PcG) for months after the first challenge infection. Sequence analysis of (PcG) DNA revealed a single amino acid change fiffering from that of Pcc. The substitution which occurs at the nt 207 position in the C-terminal 19-kDa sequence changes the amino acid glutamate into lysine. Statistically significant partial protection was observed in the immunized animals (p=0.04), despite having lower titre of antibodies ($3.3x10^3$) at the time of re-challenge. Together with sequence data, this documents the ability of recombinant MSP1inv to protect against a heterologous infection.