Quantitative Whole Genome Resequencing and Genetic Linkage Analyses Identify Genes Controlling Medically Important Phenotypes of Malaria Parasites

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Efforts are currently underway to produce a malaria vaccine, although the reality of a safe, affordable, and above all highly efficient vaccine appears to be some way off. Vaccines may be based on attenuated whole organism approaches or on sub-unit vaccines. Although attenuated sporozoite vaccines are currently being considered for development, most work to date has focused on sub-unit vaccines, as they are potentially easier to produce and distribute. The first, and potentially most crucial, step the in the development of a sub-unit vaccine is the identification of *protective* parasite antigens that may form their basis. Here, we describe an approach to identify such antigens that fuses classical genetic linkage analysis with next generation whole genome sequencing.

Using the rodent malaria parasite *Plasmodium yoelii*, we first identified parasite strains that elicit immunity in mice that is completely protective against the immunising strain, but not against a heterologous strain. Genetic crosses were then performed with these strains, and the resulting progeny grown in mice made immune to one or other of the parental strains. Whole genome sequencing at greater than 200 times coverage was then performed on both parental strains, and on the progeny pre- and post-selection, and genomic regions targeted by immune selection were identified. Three major loci under immune-selection were identified, one of which contains the major vaccine candidate gene *msp1*.

Using the same approach, we also show that major differences in growth-rate phenotypes between two *P. yoelii* strains is controlled exclusively by a gene on chromosome 13, the most likely candidate being *PyebI*.