In vitro models of macrophage activation in *Trypanosoma brucei* infection.

Pathogenesis in African trypanosome infection is associated with a dysregulation of inflammatory regulation and an over-activation of type 1 macrophage responses. This is driven in part by components of the variant surface antigen, though other factors have been implicated. The mechanisms of this process are poorly understood. The requirement for MyD88 signalling provides circumstantial evidence of TLR signalling, but no direct evidence has been presented for this. We are developing experimental platforms to define the interaction of trypanosomes with innate immune receptors, with the aim of identifying key immunomodulatory parasite components. In an *in vitro* system using murine macrophage like RAW264 cells, we demonstrate that culture adapted *T.brucei*, conditioned medium and lysate caused upregulation of inflammatory cytokine production (specifically TNF-α and IL-6). We confirmed these findings in RAW264 reporter cells that express alkaline phosphatase after PRR signalling leading to NF-ΚB activation. We also have used TLR overexpressing HEK reporter cells to demonstrate that the parasites trigger signalling via TLR4 and TLR2 *in vitro*. We will use this system to identify the ligands involved in these responses.