shown that there is an increase in soluble TREM-1 production in CM as compared to uncomplicated malaria (UM). Another study in our lab has shown that there is lower levels of endothelial progenitor cells (EPC) in CM children as compared to UM and Healthy controls (HC). Based on this result, it could be suggested that there could be an association between inflammation and microvascular damage/repair in the pathogenesis of cerebral malaria. To study this hypothesis, children between the ages of 2-12 years who are either CM, UM or HC have been recruited into the study. Samples were taken at three or four time points i.e. Day 0, (Recovery-for only CM), Day 7 and Day14. TREM-1 data and EPC data would be correlated to give a better insight into cerebral malaria pathogenesis. Findings from this study could be employed in the diagnosis of CM.

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### CLINICAL DEVELOPMENT OF A VAR2CSA-BASED PLACENTAL MALARIA VACCINE PLACMALVAC: QUANTIFYING VACCINE ANTIGEN-SPECIFIC MEMORY B & T CELL ACTIVITY IN BENINESE PRIMIGRAVIDAE

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Pregnancy associated malaria (PAM) is a major public health problem associated with poor pregnancy outcomes that commonly include maternal anemia and fetal growth alterations, whilst neonatal and infant health can also be affected. A malaria vaccine that targets the pre-erythrocytic stages of the parasite will not prevent the consequences of PAM. The identification of the parasite antigen VAR2CSA that is implicated in the pathophysiology of PAM has led to the development of a candidate vaccine by an EU-funded consortium (PlacMalVac project: German, Danish, French and Beninese Partners). The vaccine is currently under Phase I trial in Germany and Benin. As part of the PlacMalVac project, we quantified B and T cell memory responses to the VAR2CSA sub-unit vaccine candidate in a cohort of pregnant primigravid Beninese who were followed up throughout pregnancy. Clinical and parasitological data were collected every month from 37 primigravid women recruited at the beginning of their pregnancies and followed through to delivery. Mononuclear cells from peripheral blood collected on 4 occasions (first and fifth month of pregnancy, at delivery and 6 months post-delivery) were isolated and cryopreserved under liquid nitrogen. The concentrations of the cytokines IL-5, IL-6, IL-10, IL-13, IFN-γ and TNF-α produced in response to the vaccine antigen, to PPD and to PHA were quantified in supernatants of stimulated cells using cytometric bead array. The frequencies of vaccine antigen-specific antibody-secreting memory B cells were evaluated in the same cell samples using standard ELISPOT assays. Preliminary analysis shows that the profile of vaccine-specific B cell populations increased as a function of women's *Plasmodium falciparum* infection histories, whilst tetanus toxoid-specific B cell frequencies increased following tetanus vaccine boosts administered according to national guidelines. Multivariate analyses are under way to illustrate in detail the effects of *P. falciparum* infections during first pregnancies on the establishment of cellular immunological responses to the vaccine antigen.

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### THE INFLUENCE OF INHIBITORY MOLECULES ON TREG CELLS DURING *PLASMODIUM VIVAX* MALARIA

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Malaria is still considered a major helath problem worldwide, and the *Plasmodium vivax* is the most spread causative agent, with 80% of incidence in Brazil. The balance between pro- and anti-inflammatory responses is essential to limit immune response-mediated pathology and regulatory T cells (Treg) probably play an important role in this process. Recently our group demonstrated that the expression of inhibitory receptors on T cells regulates cytokines production by *P. vivax*-specific cells. The expression of one of these inhibitory receptors, the programmed death- 1 (PD-1), negatively regulates Treg function in patients chronically infected with HCV. Since the function of *P. vivax*-specific T cells is impaired due to inhibitory receptors expression, our goal is to assess the expression of these receptors on Treg upon malaria infection and to evaluate their function. Our hypothesis is that the increased expression of inhibitory receptors on Treg during *P. vivax* infection, affects the functions of these cells in regulating inflammatory responses. Peripheral blood mononuclear cells were collected from *P. vivax*–infected patients and from the same individuals after treatment, in Porto Velho-RO. Leukocytes were analyzed by flow cytometry. Our data show that *P. vivax* infection triggers an increase in the frequency of Treg cells and in the frequency of cytotoxic T lymphocyte attenuator (CTLA-4) and PD-1 expressing Treg. The expression of CTLA-4 and PD-1 on Treg was correlated with the bilirubins serum levels. Importantly, PD-1$^+$ Treg express lower levels of Forkhead Box 3 (FoxP3) than PD-1$^-$ Treg when analyzed *ex vivo* or after culture. Moreover, PD-1$^+$ Treg become able to produce IFN-γ. All together, our results indicate that malaria infection triggers the expression of PD-1 and decreases the expression of FoxP3 in Treg, phenomenum that could affect its regulatory functions.

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### ANTIBODIES TO *PLASMODIUM FALCIPARUM* APICAL MEMBRANE ANTIGEN-1 AND CIRCUMSPOROZOITE PROTEIN ARE ASSOCIATED WITH PROTECTION FROM HOSPITALIZATION AFTER SEVERE MALARIA DISEASE

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Severe malaria is a leading cause of morbidity and mortality in children. We hypothesize malaria-specific antibodies are markers of exposure and immunity; higher antibody levels will protect children against subsequent hospital sick visits and admission. A prospective cohort study was conducted at Mulago Hospital in Kampala, Uganda. Children between 18 months and 12 years with severe malaria were enrolled: cerebral malaria (CM, n=221), severe malarial anemia (SMA, n=198); and age-matched community controls (CC, n=170) and followed for one year. At enrollment, serum samples were collected and assessed for IgG antibody levels to apical membrane antigen-1 (AMA-1), circumsporozoite protein (CSP), glutamate rich protein (GLURP) and merozoite surface proteins-1 (MSP-1) using a multiplex assay. Children with SMA were the youngest, 33.5 months 41.0 months (CM), and 46.3 months (CC). Children with CM and SMA had significantly higher antibody levels for all antigens. Children with CM had higher antibody levels against malaria-specific antigens than children with SMA or CC, $p<0.05$ for all comparisons. The rate of returning sick visits for clinical malaria in the year following enrollment was 19.5% (n=43) for children with CM, 23.2% (n=46) for children with SMA, and 15.3% n=26 for CC. Higher antibody levels to AMA-1 were associated with protection from clinical malaria in CM and CC, (odds ratio