Policy brief (project no. 09-003-KU)

Execute summary

Placental malaria (PM) is an important health problem in *Plasmodium falciparum* transmission regions, with up to 125 million pregnant women at risk of malaria every year. The disease is caused by accumulation of parasite-infected erythrocytes in the placenta and a vaccine to protect pregnant women against placental malaria would save many lives each year and give a better start for even more children. In order to design a recombinant vaccine based on a 350 kDa antigen, knowledge about the molecular mechanisms behind the receptor interaction is required to direct the immune response against the specific adhesion epitope. A panel of recombinant truncated versions of VAR2CSA proteins was been constructed and testes in animal studies for antibody-reactivity against the parasite infected erythrocytes and capacity to inhibit parasite binding to the placental receptor. In this study we identified the VAR2CSA:CSA binding region which is now the leading vaccine candidate and which will be tested in the first clinical trial of a VAR2CSA-based vaccine to protect women against placental malaria.

Background

Placental malaria is caused by accumulation of parasite-infected erythrocytes in the placenta and is associated with severe maternal anemia, maternal death, preterm delivery, spontaneous abortion, stillbirth and growth retardation for the placenta and the fetus causing low birth weight. This accumulation is due to binding of the parasite expressed VAR2CSA on the surface of the IE that binds to the placental receptor chondroitin sulfate A (CSA). This makes VAR2CSA the leading protein for PM vaccine candidate but as this protein is too large to be used as vaccine, the identification epitopes responsible for induction of the anti-adhesive antibodies preventing parasite binding to CSA is key information in the vaccine development. The existing possibilities for women to protect them against PM are by using insecticide treated bed nets and intermitting preventive treatment, when accessible. However, drug resistance represents a major threat and drugs are usually first taken after first trimester of pregnancy.

This project aimed to identify an anti-adhesive vaccine that leads to induction of antibodies capable of preventing parasite binding in the placenta and thereby protect pregnant women against PM that would save many lives each year and give a better start for even more children

Methods

To identify the minimum VAR2CSA binding region required for CSA binding, we systematically produced and analyzed recombinant sub-fragments of VAR2CSA for affinity to CSA and tested the capacity of anti-adhesive antibodies obtained by animal immunization. The characterization of the specific epitopes responsible for VAR2CSA:CSA binding is important for optimal vaccine design and a crystal structure of the binding-region:CSA could give that information. Development of a monoclonal reagent against the part of VAR2CSA responsible for parasite binding to CSA would be a great benefit for the vaccine development. In this project we applied nanobody technology which is a new approach in the malaria research field, which could be a great tool to produce varied functional monoclonal reagents. Nanobodies are recombinantly produced single variable domains (VHH) from heavy-chain antibodies that are found naturally in sera of camelids and sharks in addition to the conventional hetero-tetrameric antibodies. They are the smallest naturally occurring intact antigen-binding unit and can form long, fingerlike loops that can penetrate into cavities of immunogens and are described to be able to bind cryptic antigen sites and conserved epitopes of infectious agents that are normally not recognized by conventional antibodies. Nanobodies can also be used as crystallization chaperones in crystallization and structure determination, which would give highly valuable information that can be used in the vaccine optimization.

Results

During this project we systematically analyzed recombinant sub-fragments of VAR2CSA for affinity to CSA and identified the ID1-ID2a sub-domain as the minimum binding region required for specific CSA-binding. We tested the capacity of animal induced antibodies to inhibit parasite binding to the placental receptor and found the antibodies raised against ID1-ID2a to effectively block the binding. We also included a novel approach to malaria vaccine development. In this study we used an alpaca to induce monoclonal heavy-chain antibodies targeting epitopes along the VAR2CSA, not only the immune-dominant domains of VAR2CSA, but also the N-terminal region that is responsible for VAR2CSA:CSA binding.

Conclusions and implications

In conclusion, these findings provide more insight into how to focus the vaccine to get a high efficacy of a vaccine induced response to prevent *Pf* IE binding to CSA. On the basis of these data we have received an FP7 EU grant, which will enable the first clinical trial of a VAR2CSA-based vaccine to protect women against placental malaria. This will be carried out in collaboration with the Danish ExpreS2ion Biotechnologies, the Institut de Recherche pour le Développement in France, the European Vaccine Initiative situated in Germany, the Université d'Abomey-Calavi located in Benin and the University of Tübingen in Germany. The funding from EU FP7 will enable us to perform upstream and downstream process development, the GMP production of the vaccine candidate as well as the human phase Ia and Ib clinical trials. The phase Ia clinical trial will take place in Germany led by the University of Tübingen and the phase Ib will take place in Benin and be led by the Université d'Abomey-Calavi in Benin.