**PhD PROPOSAL FOR THE**

**PASTEUR - PARIS UNIVERSITY INTERNATIONAL DOCTORAL PROGRAM**

Time for applicants to contact host laboratories: September 13 – November 2, 2017

Deadline for full application: November 13, 2017

Interviews: January 30, February 2, 2018

Start of the Ph.D.: October 1, 2018

**Title of the PhD project:** Effect of T cell immunity on antibody-dependent enhancement phenomenon in dengue viral infection

**Keywords:** dengue virus, dengue vaccine, T cell immunity, antibody-dependent enhancement, ADE, endothelial cells dysfunction

**Department:** Genome and Genetics

**Name of the lab:** Functional Genetics of Infectious Diseases

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***Doctoral school affiliation and University*:** BioSPC, Université Paris Diderot

Presentation of the laboratory and its research topics:

Our group studies the basis of human genetic susceptibility to major human pathogens and its significance to infection outcome and pathogen epidemiology. We focus on two mosquito-borne infections, malaria and dengue. We use a multidisciplinary approach combining genetic-epidemiology, vector biology, molecular genetics, genetic statistics, bioinformatics and evolutionary biology. We aim not only to identify new genes governing infection outcome, disease presentation and transmissibility, but also to understand the function and role of these genes. Over the last few years, we have focused on understanding gene-gene, gene-environment and host-pathogen interactions using human genetics/genomics and transcriptomics approaches. Studying cohorts and patient samples collected from the field, we developed new tools for genetic statistic analysis based on systems biology and we confirmed our findings by functional genetic studies both *in vitro* and in mouse models. More recently, thanks to these approaches, we have been able to deepen our understanding of the role of innate and adaptive immune responses in the development of severe dengue. Our findings have enabled us to extend into translational research with antiviral compounds testing, the development of a dengue vaccine and operational research in the field with intervention strategies. Finally, we have been solicited to implement our expertise to address two emerging infectious diseases outbreaks causing public health problems worldwide, Ebola and Zika viruses.

Description of the project:

Secondary dengue viral (DENV) infection of an individual by a different DENV serotype from the first infection often results in a more severe disease than primary infection. This is partly due to antibody-dependent enhancement phenomenon (ADE, Halstead and O'Rourke 1977; Dejnirattisai 2010). Thus, a dengue virus vaccine must induce simultaneously protective immunity against the four serotypes.

We and others have demonstrated that T cell immunity play an important role in protection against symptomatic and severe dengue. Increased frequencies of DENV-specific CD4+ and CD8+ T cells were detected in school children who subsequently experienced subclinical, compared with symptomatic secondary DENV infections ([Hatch 2011](#_ENREF_37)). A strong correlation was established between protection against severe dengue and a polyfunctional memory CD8+ T cell response with a high magnitude in healthy dengue-immune individuals ([Weiskopf 2013](#_ENREF_64)). Recently, we observed a higher activation of NK and T cells in asymptomatic dengue viral infection, indicating that different T cell populations are more proliferating and have an activated phenotype, with increased pathogen recognition, signal transduction and higher cytotoxic activity in asymptomatic DENV infected individuals compared to dengue patients (Simon-Loriere 2017).

The T cell epitopes of DENV have been mapped ([Livingston 1995](#_ENREF_43); [Kurane 1998](#_ENREF_41); [Duangchinda 2010](#_ENREF_28); [Rivino 2013](#_ENREF_55); [Weiskopf 2013](#_ENREF_64)), which reside mostly in specific regions of the non-structural proteins. Strikingly, dengue vaccine candidates have focused on induction of serotype specific neutralizing antibodies, with no cross reactivity, to avoid the ADE ([Halstead 2014](#_ENREF_35)). The T cell epitopes are lacking in most dengue vaccine candidates and the recently licensed dengue vaccine. In addition, all used *in vitro* neutralizing test as a biomarker for protective immunity and animal model to demonstrate protection against primary DENV infection.

We believe that the more efficient dengue vaccine should contain both B and T cell antigens. The better animal model for prediction of vaccine efficacy should demonstrate its protection against the effect of ADE, not only primary infection. In this proposal, we aim at obtaining **a proof of concept that Dengue vaccine containing the T cell epitopes could prevent ADE phenomenon using a mouse model**. We will optimize a mouse model that was already shown to recapitulate many aspects of human dengue disease, including vascular leakage, elevated serum cytokine levels and reduced platelet count in DENV infection (Shresta 2006; Balsitis 2010). We will use state of the art nanoparticle technology as a means for vaccination leading to highly functional DENV-specific CD4 and CD8 T cells. The immunogenicity of the NextGen dengue vaccine containing the T-cell epitopes will be tested in HLA monochain transgenic / H-2 Class I null mice (Pascolo 1997; Boucherma 2013). Since wild type or HLA class I transgenic mice are not susceptible to DENV infection, we will make them temporary interferon deficient. For the generation of ADE *in vivo*, the animals will be injected intraperitoneally with immune sera 24 hours prior to the intravenous injection of DENV serotype 1 or 2 (de Alwis 2014). ADE will be monitored by measuring systemic viral burden, fluid accumulation in visceral organs, increase in serum cytokines, and platelet depletion as well as survival rate (Balsitis 2010). Molecular mechanism of ADE-DENV induced endothelial cell dysfunction will be investigated.

References:

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Expected profile of the candidate (optional):

Be able to work on mouse model

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