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**Endocytic trafficking of antibody-opsonized mature and immature dengue virus particles**

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Dengue virus (DENV) infection currently constitutes a significant human health problem worldwide. DENV may cause disease ranging from mild febrile illness to devastating manifestations including Dengue hemorrhagic fever (DHF) and Dengue shock syndrome (DSS). Increased disease severity has been associated with pre-existing heterologous DENV antibodies, which suggests that antibodies directly influence the infectious properties of the virus. Indeed, antibodies were found to enhance viral entry into host cells and were observed to alter the antiviral immune response, leading to increased virus particle production and subsequent immune activation. Furthermore, we recently observed that anti-prM antibodies render essentially noninfectious immature particles almost as infectious as wild type virus particles and therefore may contribute to the development of DHF or DSS.

In this study, we investigate the route of cell entry of mature and immature DENV particles in the absence and presence of antibodies in macrophage-like cells. To dissect the route of cell entry we use biochemical inhibitors and molecular inhibitors in the form of dominant negative mutants to cellular proteins involved in particular endocytic pathways. Furthermore, we use triple-colored real-time fluorescence microscopy to track the entry process of single DENV particles opsonized with antibodies into the cells. For this purpose, DENV is labeled with the fluorescent probe DiD in a concentration sufficiently high to largely quench the DiD signal, but still allowing clear detection of single virus particles. Membrane fusion is evident as fluorescence de-quenching, caused by the dilution of the probe into the endosomal membrane. The endocytic trafficking behavior of antibody-opsonized mature and immature particles is assessed in macrophage-like cells co-expressing Rab5-CFP, an early endosome marker, and Rab7-YFP, a late endosome marker. The results of our study will be presented at the meeting.

Keywords: Dengue virus, Antibody-dependent enhancement, Immature viral particles, Endocytic trafficking