Differential Requirement of Cholesterol over Dengue Virus Infection Cycle

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Dengue virus is an icosahedral symmetry enveloped virus belonging to the *Flavivirus* genus, that enter into host cells via receptor-mediated endocytosis followed by fusion between the virus envelope and the endosomal membrane. Certain lipid classes seem to contribute to the fusion event by their distinct chemical structure, composition and partitioning into specific microdomains at the plasma membrane. In this way, we evaluated the requirement of specific lipids to the fusion reaction of dengue virus serotype 2 (DENV-2) using a liposomal model (ANTS/DPX) that allows the investigation of content mixing between distinct compartments. DENV-2 fused efficiently with receptor-free liposomes consisting of phosphatidylcholine, phosphatidylethanolamine, cholesterol and sphingomyelin, suggesting that receptor interaction is not crucial for fusion. However, cholesterol absence in this model blocked the fusion reaction, while lack of sphingomyelin was indifferent to the process. Given these findings, we next evaluated the effect of cholesterol depletion from cell plasma membrane by methyl-ß-cyclodextrin on virus infection efficiency, where we also observed a requirement of this lipid to DENV-2 infection process. Paradoxically, despite this cholesterol requirement for entry, DENV-2 infection seems to modulate negatively host cell cholesterol synthesis, as assessed through thin-layer chromatography. Furthermore, virus infection seems to decrease cell membrane fluidity, as observed through two-photon excitation fluorescence microscopy using the lipophilic fluorescent probe laurdan. Our results suggest that DENV-2 shows a differential requirement of host cell cholesterol over its infection cycle.

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