The Principles of Viral Lipid Uptake: A Lipidomics Approach

Julio L. Sampaio, L. Kalvodova, K. Simons, A. Shevchenko, MPI-CBG
Dresden, Germany

How enveloped viruses include lipids into their membrane during budding from the infected host cells remains poorly understood. Viral proteins exclude host cell proteins from the newly formed capsule. Moreover, some viruses when co-infected with others don't "share" the same membrane space, which results in the formation of 2 individual populations of viruses without any phenotypic mixing. This observation suggests that these viruses could bud from specific membrane domains a. k. a lipid rafts.

To analyze this issue, we took advantage of the fact that baby hamster kidney cells can be infected by two different viruses, vesicular stomatitis virus, and Semliki Forrest virus from the *Rhabdoviridae and Togaviridae* families, respectively. Importantly, their viral proteins don't co-patch in a co-infection experiment making them good candidates for specific lipid uptake during budding. We purified the host plasma membrane and the two different viruses after exit from the host cell. We analyzed the lipid composition of these membranes by quantitative shotgun mass spectrometry. The combination of optimized lipid extraction with high resolution mass spectrometers enabled us to quantify the 13 major lipid classes comprising 159 individual lipid species from minute lipid amounts (< 3 µg of lipid) coming from complex biological preparations. Importantly, for the first time we include in the analysis the absolute quantification of the ganglioside GM3, which turned out to be a major constituent of the plasma membrane and viral envelopes.

We observed that the lipid compositions of both of these otherwise structurally different viruses are virtually identical. When the viral lipid composition was compared to the lipid composition of the plasma membrane we conclude that these viruses exert small, yet significant specificity in engaging lipids into their envelopes, which seem to be driven by the viruses curvature. PM is considered to be a plane bilayer while VSV is mildly curved (bullet shaped virus 180x70 nm) while SFV is highly curved (spherical virus with 50 nm in diameter). PS is predominantly enriched in the inner leaflet of the PM, while PC and SM are in the outer leaflet. During the budding, the curvature is generated and PS species with bulky fatty acids (di-unsaturated) are sorted to the budding site of both viruses. The extreme curvature of SFV also sorts SM and saturated and mono-unsaturated PC species to the budding region.