Crossing the barrier: Understanding the life cycle of membrane-containing phages at molecular resolution

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Bacteriophages are the most common biological entities found in every biosphere. Bacteriophages can be classified based on the nucleic acid composition, capsid symmetry, and presence or absence of a lipidic membrane. Unlike mammalian viruses, relatively few known bacteriophages have an inner or outer membrane. This is partly due to the limitations of current isolation protocols, which involve organic solvents. Although membrane-containing phages were identified over 50 years ago, the detailed mechanisms of phage entry, mature virion packaging, membrane acquisition from the host, and envelope decoration with spike proteins remain poorly understood.

In this study, we elucidated the molecular and structural intricacies of the lifecycle of enveloped phages (EnvB) at unprecedented resolution by combining electron cryotomography (cryo-ET) with genetics and fluorescence microscopy. To achieve detailed visualisation of the infection process, we used multiple 3D segmentation software, enabling high-resolution insights into each stage of the phage-host interaction. We captured the nuances of attachment, entry and the assembly of mature EnvB phages. Using cryo-ET, we revealed that EnvB phage undergoes striking morphological changes during its attachment to the host cell, followed by membrane fusion. We captured the phage engulfment by macropinocytosis, formation of viral factories and bacterial "endomembrane-like" structures. Additionally, we have shed light on the role of EnvB phage proteins in facilitating the lytic cycle within the host. These findings provide significant molecular insights into the infection cycle of EnvB phage and uncover new parallels between EnvB phage and mammalian/eukaryotic viruses.