Thesis Title: Investigation of the structural dynamics of non-enveloped virus disassembly using Flock House virus as a model system

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Abstract

The stability of icosahedral virus capsids is essential for providing maximum protection to the genome in transit; however, successful infection requires eventual disassembly of the capsid and release of packaged genome within the cell. A comprehensive understanding of how stable and uniform icosahedral particles disassemble within host cells remains elusive, which indicates the complexities involved in either isolating unstable intermediate structures from infected cells or in establishing a comprehensive and dependable method for generating intermediates *in vitro*. We utilized incremental heating to isolate and characterize two disassembly intermediates – "eluted particles" and "puffed particles" of an insect nodavirus, Flock House virus (FHV). Cryo-electron microscopy and single-particle 3D reconstruction of the eluted particles indicated that disassembly-related conformational alterations are minimally global and largely local, leading to asymmetry in the particle. Asymmetric reconstruction, primarily utilized in this study reconstructed unique 3D density maps of the intermediates, which revealed a global loss of γ peptide and portions of N- and C-termini of β capsid protein of FHV, along with a series of local structural changes that were confined to a specific region on the intermediates. Notably, the 2-fold symmetry axis was found to be the weak link and the probable site for genome release. Targeting of the virus local symmetry was also mapped to the encapsidated RNAs, which showed a directional release from the capsid. Further, intriguing differences in the disassembly pattern of a non-infectious, maturation defective FHV variant suggested an important link between the capsid maturation and disassembly. Our study presents a comprehensive structural analysis of the sequence of programmed conformational changes during FHV uncoating, and provides insight into the mechanism of non-enveloped virus disassembly in general.