Oncolytic Seneca Valley Virus structure and structural insights into receptor specificity

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Oncolytic viruses (OVs) are replication competent agents that selectively target cancer cells. Seneca Valley Virus (SVV) is a newly-discovered oncolytic picornavirus which is classified as the sole member in the genus *Senecavirus*. SVV strain 001 has currently completed Phase I and Phase II clinical trials in pediatric solid tumors and small-cell lung cancer, respectively. Similar to other picornaviruses, SVV forms naturally occurring empty capsids (without genome), known as procapsids, which share the same antigenicity as full virions. Understanding the formation and structure of SVV procapsids could give insights into how to exploit them as virus-like particles (VLPs) for targeted in vivo drug delivery for cancer treatment. Recently, we identified the Anthrax Toxin Receptor 1 (ANTXR1), a membrane protein overexpressed in ~60% of types of cancer, as the high-affinity cellular receptor for SVV in cancer cells[1]. However, the high-resolution information on SVV-ANTXR1 interaction sites remained poorly characterized, thereby hampering the potential to develop SVV mutant in future oncovirotherapy.

Here, we present how we purified SVV full capsids and procapsids using density gradient ultracentrifugation and used cryo-electron microscopy (cryo-EM) to solve the structures of full capsid, procapsid and full capsid-ANTXR1 complex to resolutions of 3.29 Å, 5.9 Å, and 3.8 Å respectively[2,3]. Our results show that both full capsids and procapsids have a similar external structure, while on the interior the main differences were the missing genomic RNA in the procapsid and a disordered VP1 region. Structural protein analysis in SDS-PAGE revealed the presence of capsid proteins VP1-VP4 in both full capsids and procapsids. We also show that a cage of RNA serves to stabilize the inside surface of the full capsid, thereby making it more acid stable. In SVV-ANTXR1 complex, ANTXR1 decorates the outer surface of the SVV capsid and interacts with the surface-exposed BC loop and loop II of VP1, “the puff” of VP2 and “the knob” of VP3. Comparison of the receptor-bound capsid structure with the native capsid structure reveals that receptor binding induces minor conformational changes in SVV capsid structure, suggesting the role of ANTXR1 as an attachment receptor. Our results demonstrate that the capsid footprint on the receptor is not conserved in anthrax toxin receptor 2 (ANTXR2), thereby providing a molecular mechanism for explaining the exquisite selectivity of SVV for ANTXR1.

Findings from this study lay the foundation for the modification of the SVV procapsid to develop it for targeted *in vivo* delivery of therapeutics and to develop potent SVV mutants with specific cancer tropism.

References

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