Poster 28

Molecular Modeling of AAV Capsid-capsid Interaction Under Different Salt Concentrations, and Surfactants to investigate the aggregation

Authors: Leila Sharifi (1), Tibo Duran (1), Shivangi Naik (2), Willow DiLuzio (2), Arani Chanda (2) and Bodhisattwa Chaudhuri (1,3)
Presenting Author: Leila Sharifi
Affiliation: (1) Pharmaceutical Sciences, University of Connecticut, Storrs, CT, (2) Sarepta Therapeutics, Cambridge, MA, (3) Chemical and Biochemical Engineering, University of Connecticut, Storrs, CT.
Corresponding Author's email: bodhi.chaudhuri@uconn.edu

Purpose

Adeno-associated virus (AAV) is a non-enveloped virus with approximately 26 nm diameter, which exhibits great potential as a gene delivery tool. However, its manufacturing has challenges such as lower yields, and moderate purity, attributed to degradation during various stages of production. This degradation occurs potentially via aggregation at low ionic strength and insufficient surfactant concentration. These mechanisms of degradation remain poorly understood, and there is a lack of a systematic approach to formulation development. To address this, a coarse-grained molecular dynamics (CG-MD) simulations approach was utilized to understand the molecular level of capsid-capsid interaction under different types and concentrations of salts and surfactants. The model was validated with experimental data.

Methods

CG-MD simulations were performed using the GROMACS package with MARTINI force fields. AAV8 (PDB: 6v12) with 2.2kb single-strand DNA was selected as the model viral capsid. Energy minimization used the steepest descent algorithm, followed by a 10 ns equilibration step. Production runs were performed using the Nosé−Hoover thermostat and the Parrinello−Rahman barostat at 1 bar pressure for each 20-fs time step in an isothermal-isobaric ensemble.

Results

We investigated the effect of different types and concentrations of salts on AAV vector aggregation. Our results showed that NaCl and $MgCl_2$ salts at various concentrations could prevent aggregation to some extent. AAV vector aggregation was influenced by the ionic strength of the solution, and NaCl salts were effective at preventing aggregation when present at a concentration above 0.05 M. Additionally, surfactants such as poloxamer 188 at concentrations ranging from 0.001% to 0.01% (w/v) were tested, and our findings suggest that the type and concentration of surfactants are critical factors that affect AAV vector aggregation.

Conclusion

Our developed computational model, with a microsecond timescale, evaluated the behavior of different salts and surfactants with varying concentrations to understand the factors preventing AAV8 aggregation. This analysis provided insights that would be difficult to obtain from experimental studies. The CG-MD simulations proved powerful in simulating macromolecular complexes and guiding the interpretation, and direction of experiments. Our simulations provided important insights into AAV vector formulation development, which could lead to the development of more efficient and stable gene delivery systems.

Keywords: Adeno-associated viruses, molecular dynamics, capsid aggregation, protein interaction, ssDNA, salt, surfactant.