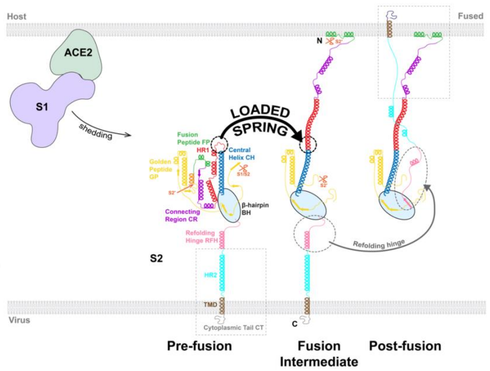
Refolding mechanism of the SARS-CoV-2 Spike protein for cell entry

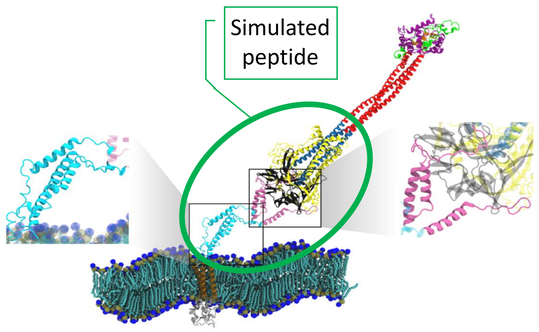
Stephanie Morales[1], Hongkang Zhu[1], Dong An[1], Tianyi Zhu[1], Ben O'Shaughnessy[1]

[1]Department of Chemical Engineering, Columbia University, NY 10027, USA

INTRODUCTION

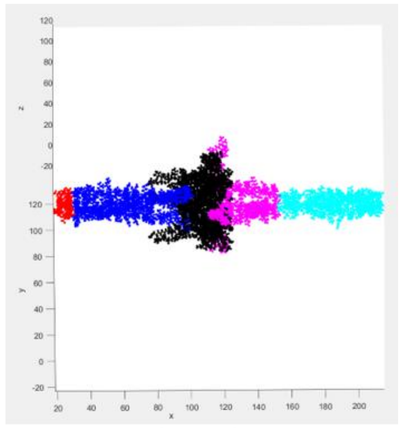
- S1 subunit binds to the ACE2 receptor and the S2 dissociates from S1
- The S2 subunit straightens itself into the long Fusion Intermediate, with the fusion peptides at the tip that capture the host cell membrane
- Then the S2 refolds to bring the host cell membrane close to the virus membrane, triggering membrane fusion and achieving cel entry
- During the Fusion Intermediate, we hypothesize that the refolding starts when the RFH and HR2 domains become destabilized





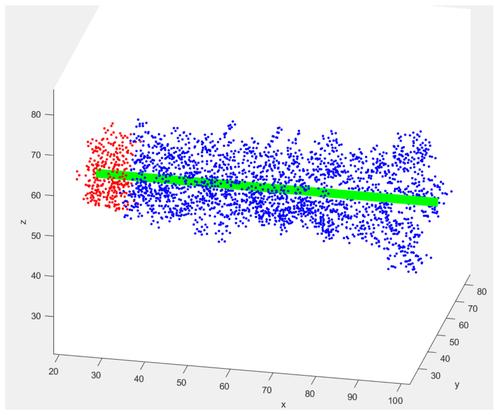
R. Su, J. Zeng, B. O'Shaughnessy, Host cell membrane capture by the SARS CoV-2 spike protein fusion intermediate. *bioRxiv* https://doi.org/10.1101/2021.04.09.439051 (2021).

RESULTS



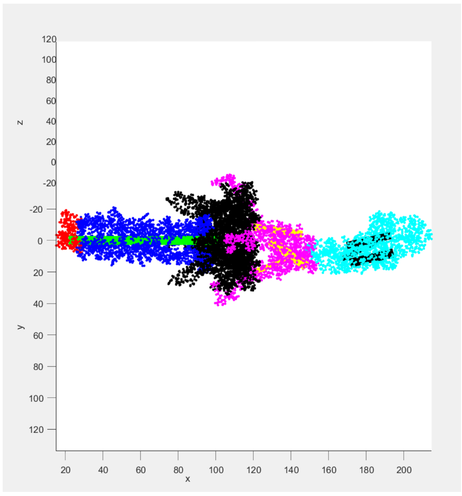
Initial condition

- Angles taken in relation to the backbone because it is rigid
- At 30 ns, the RFH and the HR2 peptide become unstable
- Used a secondary structure algorithm to find the parts of the peptides that are alpha helical after 30 ns
- Having the RFH and the HR2 destabilized, the S Protein can refold and bring the host and the virus membrane together

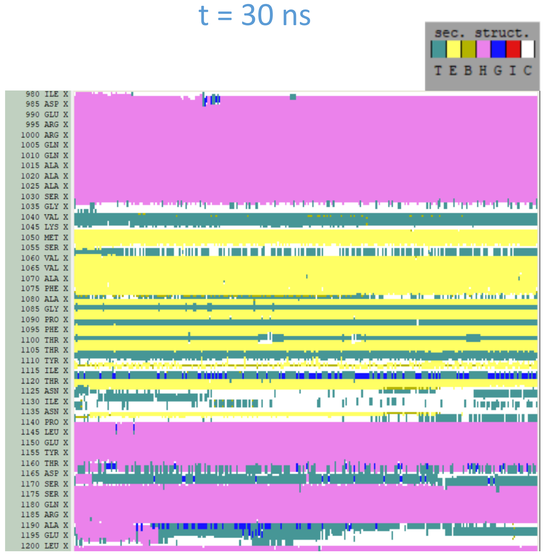


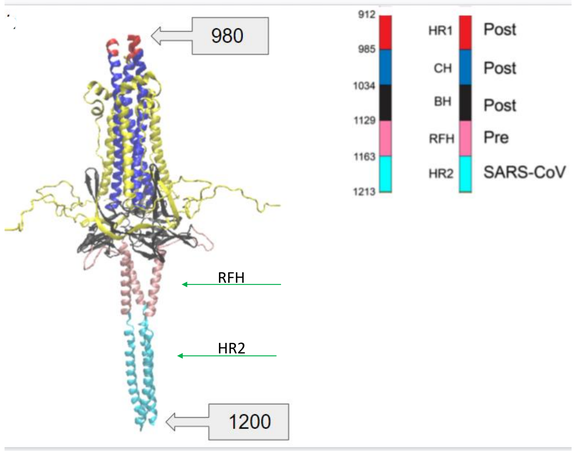
Fitting the orientations of alpha helices

t = 0 ns	θ_1	θ_2	θ_3	$\Phi_{1 \text{ to } 2}$	$\Phi_{2 \text{ to } 3}$	$\Phi_{1 \text{ to } 3}$
RFH	36°	36°	36°	38°	22°	61°
HR2	5°	6°	7°	2°	28°	30°
t = 15 ns	θ_1	θ_2	θ_3	$\Phi_{1 \text{ to } 2}$	$\Phi_{2 \text{ to } 3}$	$\Phi_{1 \text{ to } 3}$
RFH	27°	19°	44°	8°	75°	67°
HR2	28°	15°	21°	25°	21°	46°



t = 30 ns

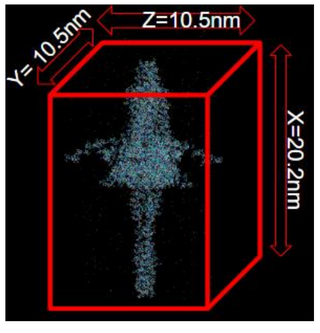




Simulated a smaller portion of the protein (simulated by Su et al.,) for acceleration

METHOD

- Data analysis by MATLAB
- Molecular Dynamics
  - VMD
  - CHARMM-GUI
  - GROMACS



Chose an appropriate water box

CONCLUSIONS

- The backbone and parts of RFH and HR2 remain alpha helical
- In the initial condition, the HR2 helices are parallel to the backbone
- The orientations of RFH and HR2 change significantly after 30 ns of simulation
- Reorientation of the RFH and the HR2 helices are early indicators of destabilization and refolding of the Fusion Intermediate

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