UNDER THE LENS

Simulations of the spike: molecular dynamics and SARS-CoV-2

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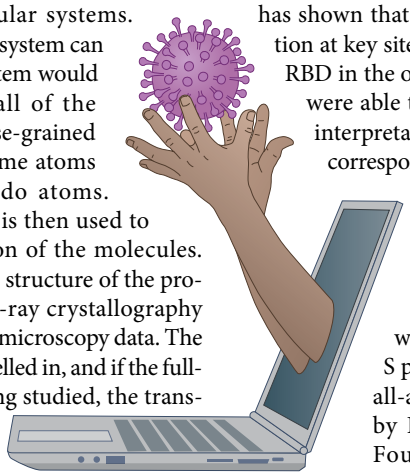
This month's Under the Lens discusses a few of the growing number of recent molecular simulation studies that have made substantial contributions towards our mechanistic understanding of the spike protein of SARS-CoV-2.

Antibody development to SARS-CoV-2, the causative agent of COVID-19, has largely revolved around the viral spike (S) glycoprotein, which mediates host cell entry by binding to a cell surface receptor, angiotensin-converting enzyme 2 (ACE2). The S protein is a trimeric class I fusion protein, in which each monomer is composed of two subunits: S1 and S2. The receptor-binding domain (RBD), where interaction with ACE2 occurs, is located within subunit S1. Proteolytic cleavage at the S1–S2 site exposes the S2 site, liberating the fusion peptide and initiating the process of infection. The details of the conformations, orientations and dynamics of the many glycans that decorate the surface of the S protein are not attainable from structural studies.

Molecular dynamics is a computational technique that enables prediction of the time evolution of molecular systems. The resolution of the system can vary; an all-atom system would explicitly include all of the atoms, whereas coarse-grained systems combine some atoms together into pseudo atoms. Classical mechanics is then used to propagate the motion of the molecules. For the S protein, the structure of the protein is taken from X-ray crystallography and/or cryo-electron microscopy data. The glycans are then modelled in, and if the full-length protein is being studied, the transmembrane domain is embedded into a model membrane. Many days of simulations of such systems will typically yield trajectories that represent microseconds of molecular motion.



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In a recent study, Amaro and co-workers[1] were able to identify a unique functional role of the *N*-linked glycans at sites neighbouring the RBD by modulating the conformational dynamics of the S protein and its impact on ACE2 binding. This was achieved through all-atom simulations of the glycosylated, full-length S protein on timescales of multiple microseconds. The extended simulations also enabled characterization of the vulnerabilities of the entire glycan shield of the S protein, which may inform future development of vaccines and antiviral agents. In subsequent work, a team lead by Amaro and Chong used a weighted ensemble approach to perform molecular dynamic simulations of the S protein head, in a study that revealed that a specific glycan along with three charged residues facilitate opening of the RBD, required for binding to ACE2 (REF.[2]). In both studies, the predictions from simulations were corroborated by experimental studies. More recently, a detailed molecular dynamics simulation study of the S protein glycans by Fadda and co-workers[3] has shown that the nature of the glycosylation at key sites impacts the stability of the RBD in the open state. Furthermore, they were able to provide a molecular-level interpretation for increased infectivity corresponding to a recent loss of glycosylation from a key site owing to viral evolution. These predictions were experimentally validated in later work.

Details of the flexibility within the stalk region of the S protein were revealed by large all-atom simulations performed by Hummer and co-workers[4]. Four glycosylated S proteins anchored into a patch of viral membrane were simulated in a system containing more than four million atoms. The results showed three distinct hinge regions, which provide the head region of the protein with substantial orientational flexibility. The authors speculated that the conformational freedom provided by the hinges may interfere with antibodies gaining access to the stalk region as well as facilitate engagement of the S protein with host membranes. Although for a system of such a large size, and as highlighted by the authors themselves, the simulations (2.5 microseconds) may not have sampled all of the conformational space, there was good agreement with cryo-electron tomography data.

In all of the cases described here, the S protein has been simulated with its glycans, for multiple microseconds, taking input from multiple sources of experimental data to set up the simulations to reveal key details with a fundamental and likely therapeutic impact. In doing so, the authors have shown that molecular dynamics simulations are an indispensable tool for mechanistic studies of viruses, and with availability of growing computational power and the advent of enhanced sampling methods, their contributions to this field will continue to grow.

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1. Casalino, L. et al. Beyond shielding: the roles of glycans in the SARS-CoV-2 spike protein. *ACS Cent. Sci.* **6**, 1722–1734 (2020).
2. Sztain, T. et al. A glycan gate controls opening of the SARS-CoV-2 spike protein. *Nat. Chem.* **13**, 963–968 (2021).
3. Harbison, A. M. et al. Fine-tuning the spike: role of the nature and topology of the glycan shield in the structure and dynamics of the SARS-CoV-2 S. *Chem. Sci.* **13**, 386–395 (2022).
4. Turoňová, B. et al. In situ structural analysis of SARS-CoV-2 spike reveals flexibility mediated by three hinges. *Science* **370**, 203–208 (2020).

Competing interests
The authors declare no competing interests.