NCCAT GUP1: Structure determination of maturation intermediates of the SARS-CoV-2 spike

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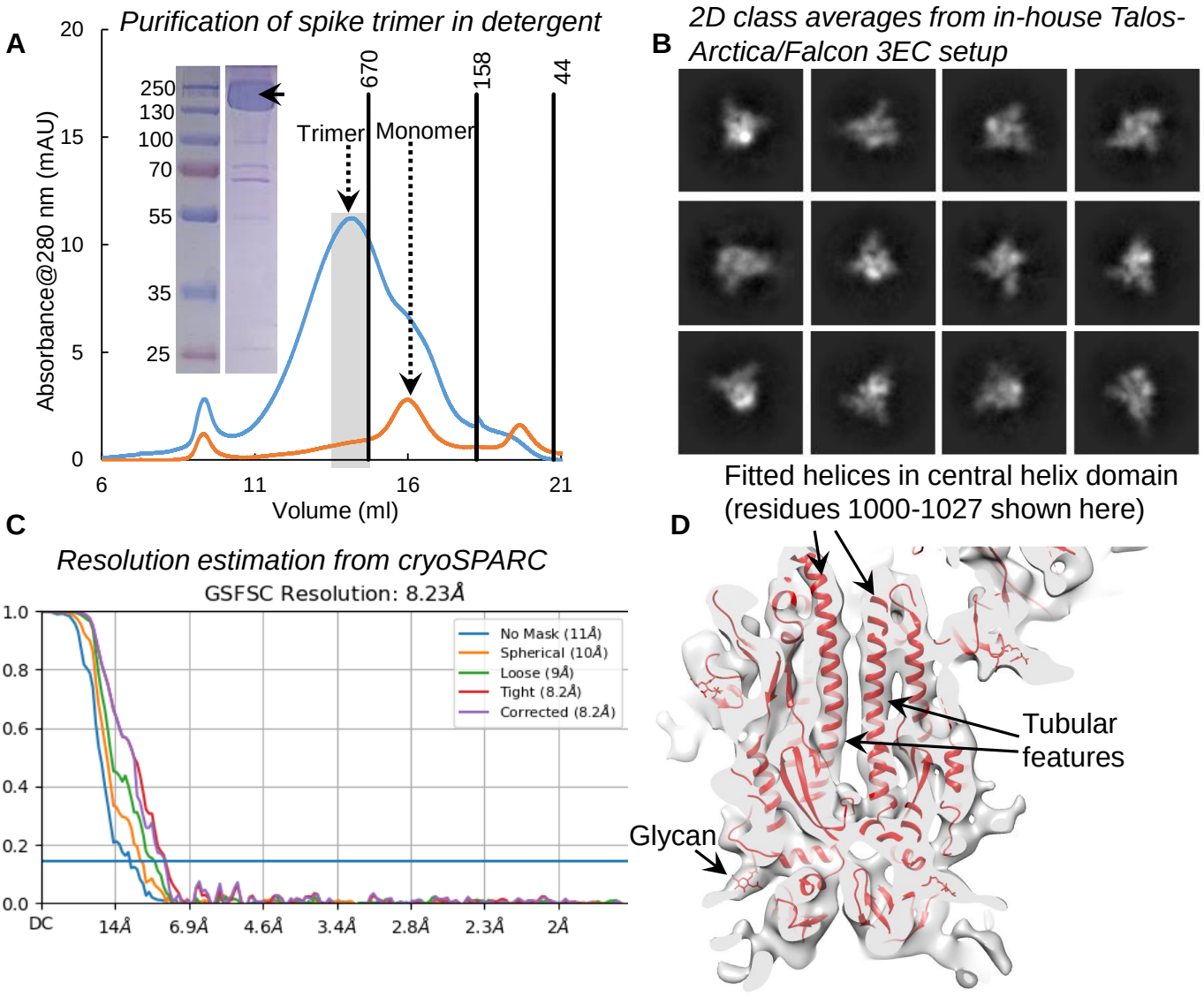
Supplemental Information to Establish Feasibility



Legend: Preliminary data to establish feasibility of our proposed project on SARS-CoV-2 spike. (**A**) Chromatogram from Superose 6 10/300 Increase size-exclusion column showing the trimeric spike peak in gray. This is the spike construct that displays immature-state glycans. The inset shows the purified spike trimer band on denaturing and reducing SDS-PAGE. As a control, we generated a spike monomer (orange trace) by reducing the detergent concentration. For reference, we have included the positions of calibration standards (670, 158, and 44 kDa). (**B**) Selected class averages of spike trimer particles selected from blob-picking in cryoSPARC. The data were collected on our in-house 200 keV Talos-Arctica/Falcon 3EC setup. (**C**) FSC curves showing the gold-standard resolution estimate at 8.2Å for ~8,600 particles. (**D**) The 8.2Å resolution cryoEM map of the spike trimer shows tubular features into which helices from the central helix domain are fitted. This was done in Chimera by fitting the published spike trimer structure, PDB ID 6XR8 (shown in red) in this cryoEM map. This shows that the map is overall consistent with the spike structure at the reported resolution. At this resolution, a glycan site is prominent as a knob-like feature on the protein surface. *The grids from the batch that yielded this 8.2Å resolution reconstruction of the spike trimer have been saved for data collection in NCCAT.*