

Preliminary data

Purified NST-BRIL-sAB complex at 5mg/ml was applied onto glow-discharged 300 mesh grids (Quantifoil R 2.0/2.0) and plung-frozen into liquid ethane using a FEI Vitrobot (Thermo Fisher Scientific). Data were collected on a Titan Krios electron microscope operated at 300 kV using EPU (16) with a nominal magnification of 135,000x (resulting in a calibrated physical pixel size of 0.67 Å/pixel) and a defocus range of 1.5-3.5 μm . The images were recorded on a K3 summit electron direct detector (Gatan, Pleasanton, CA) in counting mode at the end of a GIF-Quantum energy filter (Gatan, Pleasanton, CA) operated with a slit width of 20 eV. A dose rate of 15.84 $\text{e}^-/\text{pix}/\text{s}$, generating 48 movie frames with a total dose of 58.84 electrons per \AA^2 .

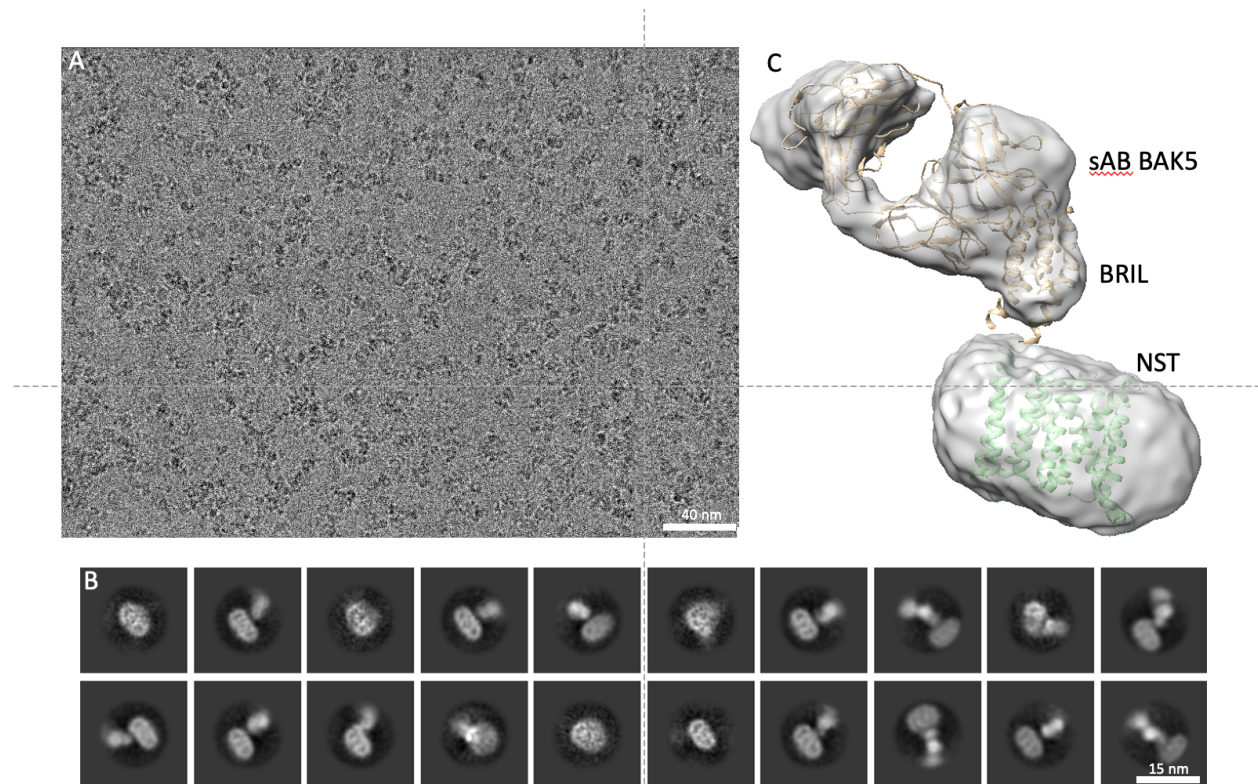


Figure: Cryo-EM single particle reconstruction NST-BRIL-sAB complex **a** A representative cryo-EM micrograph of NST-BRIL-sAB complex reconstituted in PMALC8, **b** representative 2D class averages and **c** cryo-EM map docked with NST (pdb:QSK) and BRIL-sAB BAK5 (pdb:6USF).