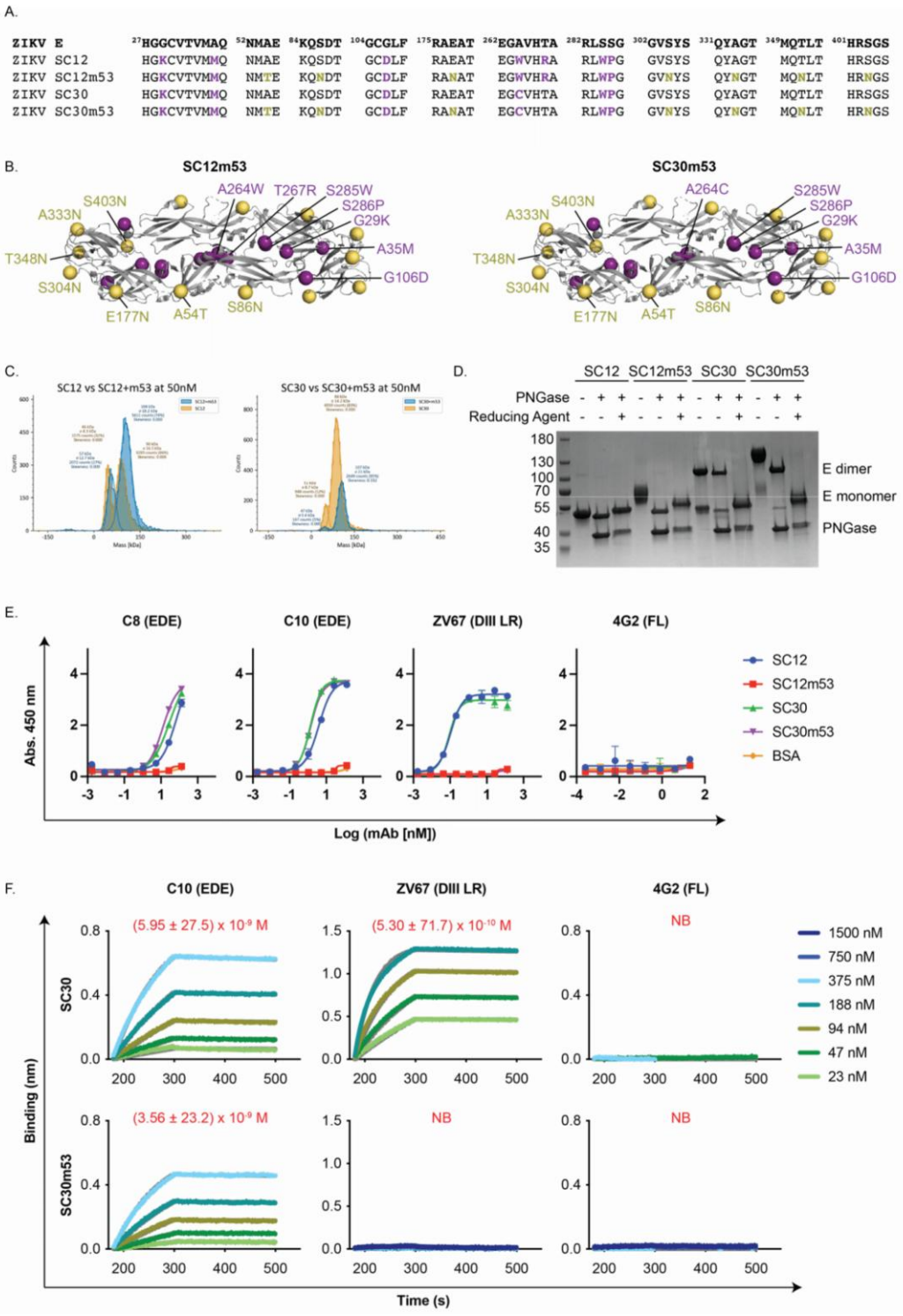

Figure 1

Biochemical characterization of SC ZIKV E variants.

(A) Sequences of ZIKV E (PDB 5LBS), and stabilizing combination (SC) ZIKV E variants (SC12, SC12m53, SC30, SC30m53). Positions of installed EDE-binding positive glycan mutations (**yellow**), and stabilizing combination mutations (**purple**) are indicated.
(B) Model of SC12m53 and SC30m35 with positions of installed EDE-binding positive glycan positions (**yellow**) and stabilizing combination mutations (**purple**).
(C) Mass photometry showing comparison between expected molecular weights of sE proteins to molar mass of species and distribution of oligometric states in solution of SC12, SC12m53, SC30, and SC30m35.
(D) SDS PAGE mobility gel shift assay of SC12, SC12m53, SC30, and SC30m35 under reducing and non-reducing conditions with and without PNGase.
(E) Binding reactivity of EDE mAbs C8 and C10, and non-EDE mAbs ZV-67 and 4G2 to SC12, SC12m53, SC30, and SC30m35 determined by ELISA. Representative data from two independent experiments completed in duplicate are plotted as the (average absorbance at 450 nm) ± SD.
(F) Reactivity profiles for SC30, and SC30m35 to EDE mAb C10, and non-EDE mAbs ZV-67 and 4G2 as determined by biolayer interferometry (BLI). Shown are representative data from two independent experiments with consistent results. Apparent KD (KDapp) values are indicated for each sensorgram, weak binding (WB), no binding (NB).

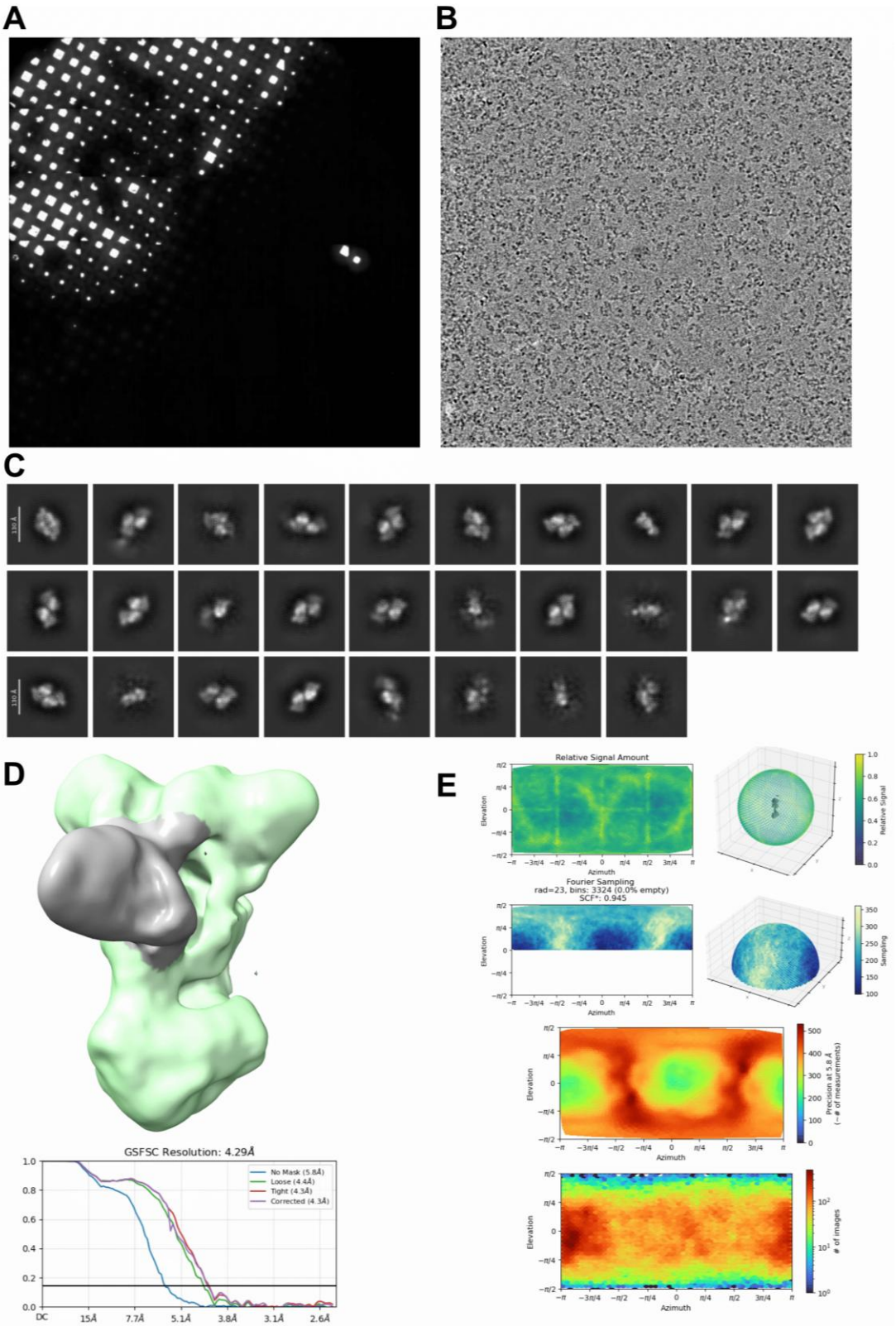

Figure 2

Data collection analysis of a Tundra dataset of (1200 micrographs after manual curation) collected on SC30m53: Fab C10 complex sample.

(A) Atlas of the grid SC30m53: Fab C10.
(B) Representative micrograph.
(C) Selected 2D classes from data processing with cryoSPARC.
(D) Top panel: Preliminary map, in C1 symmetry, obtained with ab-initio reconstruction followed by heterogeneous refinement, and 2 rounds of non-homogeneous refinement, local CTF refinement, global CTF refinement and finished with non-homogeneous refinement. The number of final particles was 200334. Bottom panel: Fourier Shell Correlation (FSC) curves calculated between the two half-maps in the last non-homogeneous refinement using cryoSPARC. The grey color indicates the volume region that we expect to belong to the fab and in green the volume of the Zika sE dimer.
(E) Orientation diagnostic run on cryoSPARC shows low presence of preferred orientation. Panel 1 (Top): Half-map correlation as function of viewing direction shows that relative signal is homogeneous and few direction regions have low signal. Panel 2: Sample compensation factor (SCF) measurements suggest a good sample distribution with an SCF score of 0.945. Posterior precision directional distribution (panel 3) and viewing direction distribution plot (panel 4) suggests low orientation preference.