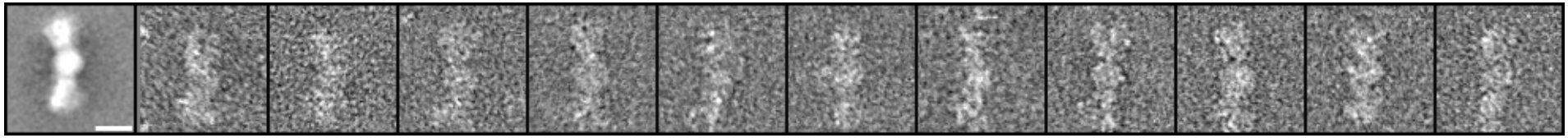

Figure S1. Rosettes of SOSIP.681. Reference-free class average of a rosette observed with SOSIP.681 stained with 2% Uranyl-formate. The first image is the class average and following images are representative raw particles observed in this average. The white bar represents 100Å, images were collected at 100,000x magnification.

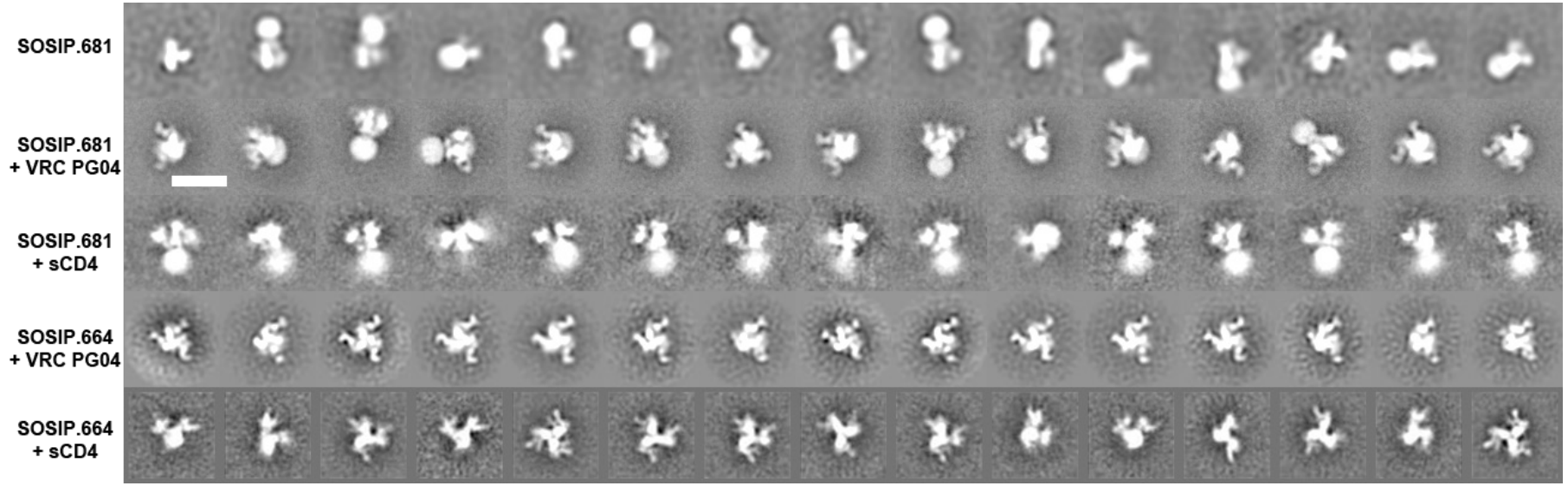

Figure S2. Reference free class averages of selected constructs. Reference free class averages exhibiting different particle orientations. The range of particle orientations was obtained using our tilted data collection strategy. Each pixel is 4.36Å, and the white bar measures 200Å.

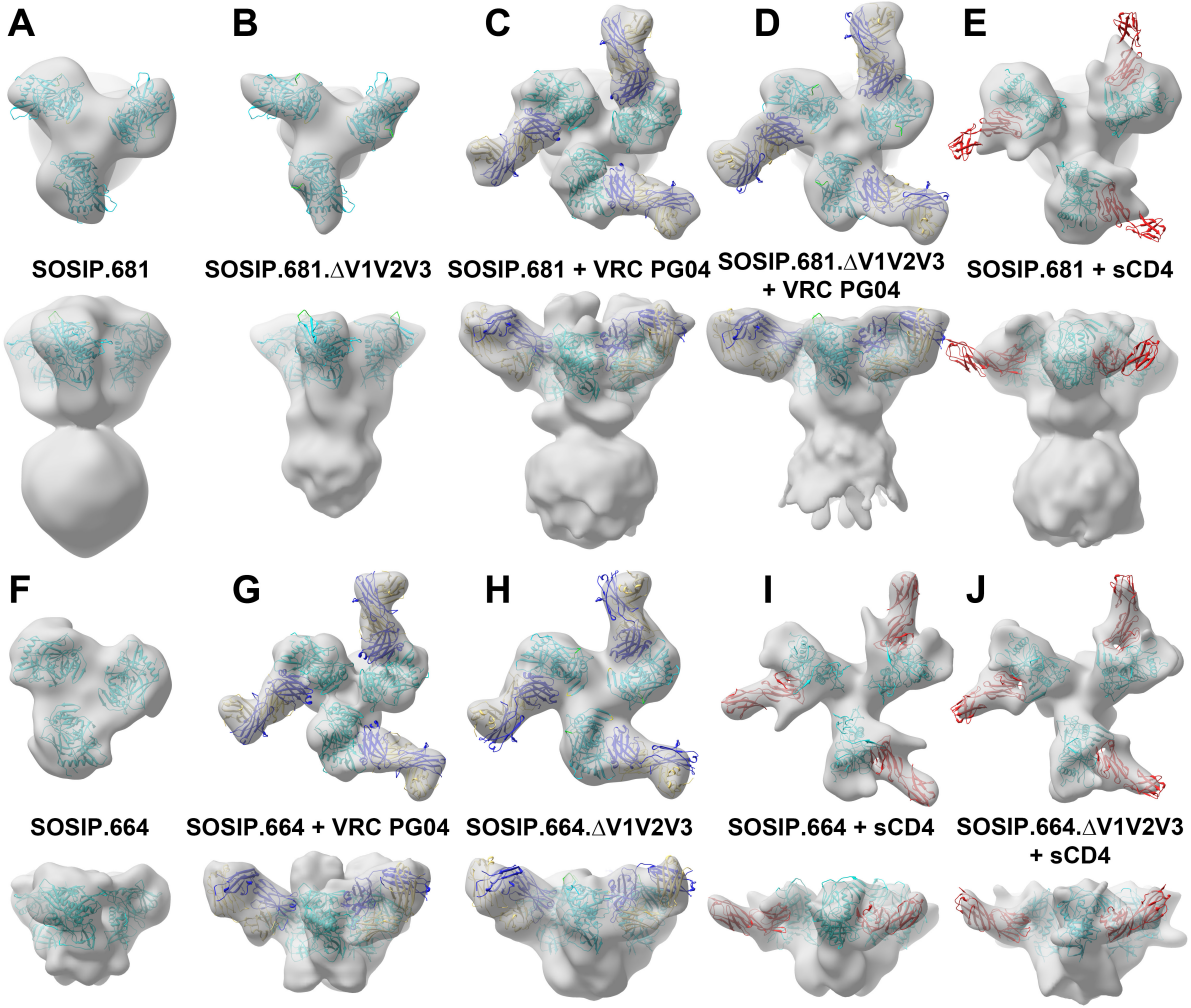

Figure S3. Crystal structures fitted into the SOSIP.681 and SOSIP.664 image reconstructions. A surface representation of the SOSIP image reconstructions with the crystal structures docked using an automated procedure (see methods and materials). A) SOSIP.681 (PDB entry: 3TIH), B) SOSIP.681.ΔV1V2V3 (PDB entry: 3TIH), C) SOSIP.681 + VRC PG04 (PDB entry: 3SE9), D) SOSIP.681.ΔV1V2V3 + VRC PG04 (PDB entry: 3SE9), E) SOSIP.681 + sCD4 (PDB entry: 1GC1). Coloring is as follows: gp120core (cyan), VRC PG04 heavy chain (blue), VRC-PG04 light chain (light yellow), sCD4 (red). F) SOSIP.664 (PDB entry: 3TIH), G) SOSIP.664 + VRC PG04 (PDB entry: 3SE9), H) SOSIP.664.ΔV1V2V3 + VRC-PG04 (PDB entry: 3SE9), I) SOSIP.664 + sCD4 (PDB entry: 1GC1), and J) SOSIP.664.ΔV1V2V3 + sCD4 (PDB entry: 1GC1). The image reconstructions were binned by 2 in UCSF Chimera.

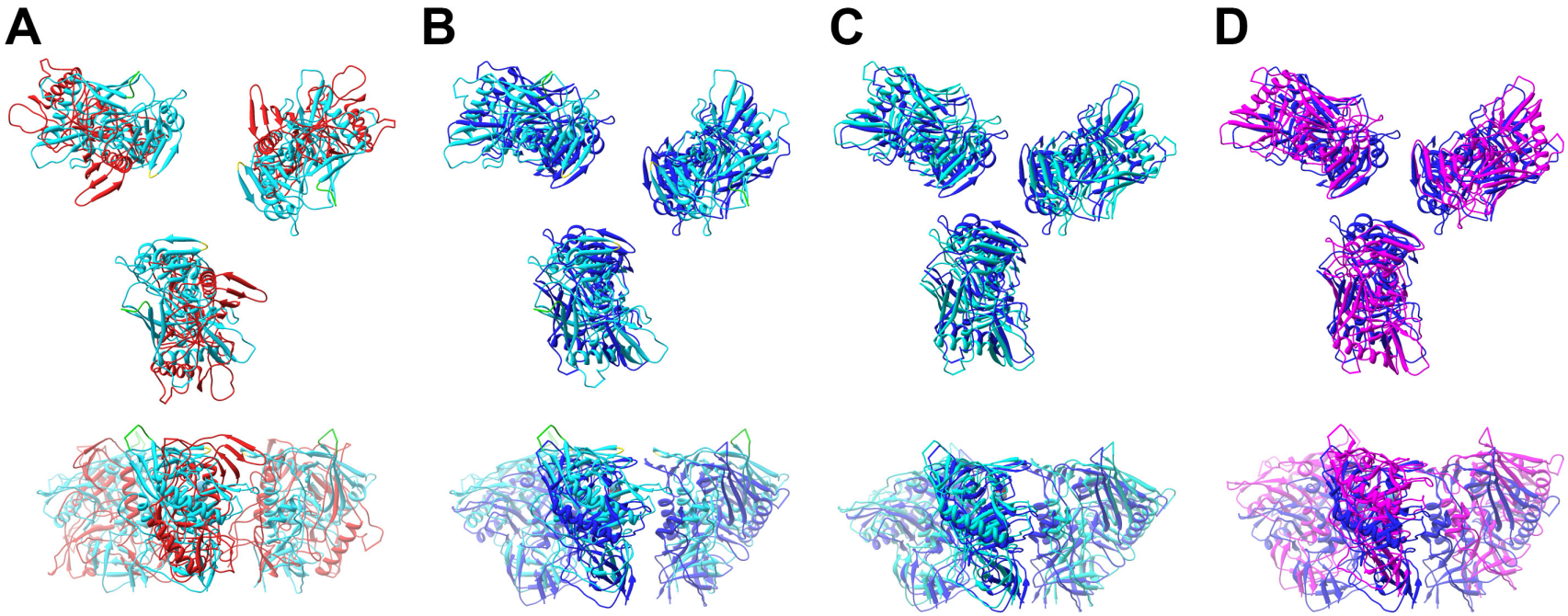

Figure S4. Positional and rotational changes in gp120. Ribbon representation of the crystal structures of gp120 fitted into the SOSIP image reconstructions. Note, the ligands (sCD4 and VRC PG04) were computationally removed after the fitting to generate the figures. Additionally, the gp120 from PDB entry 3TIH was overlaid onto the fitted structures to generate an identical gp120 structure to simplify the comparison. A) comparison of SOSIP.681 (cyan, PDB entry: 3TIH) and SOSIP.681 + sCD4 (red, PDB entry: 1GC1), B) comparison of SOSIP.681 (cyan, PDB entry: 3TIH) and SOSIP.681 + VRC PG04 (blue, PDB entry 3SE9), C) SOSIP.681 + VRC PG04 (cyan, PDB entry 3SE9) and SOSIP.664 + VRC PG04 (blue, PDB entry: 3SE9), and D) SOSIP.664 + VRC PG04 (blue, PDB entry: 3SE9) and SOSIP.664.ΔV1V2V3 + VRC PG04 (magenta, PDB entry 3SE9).

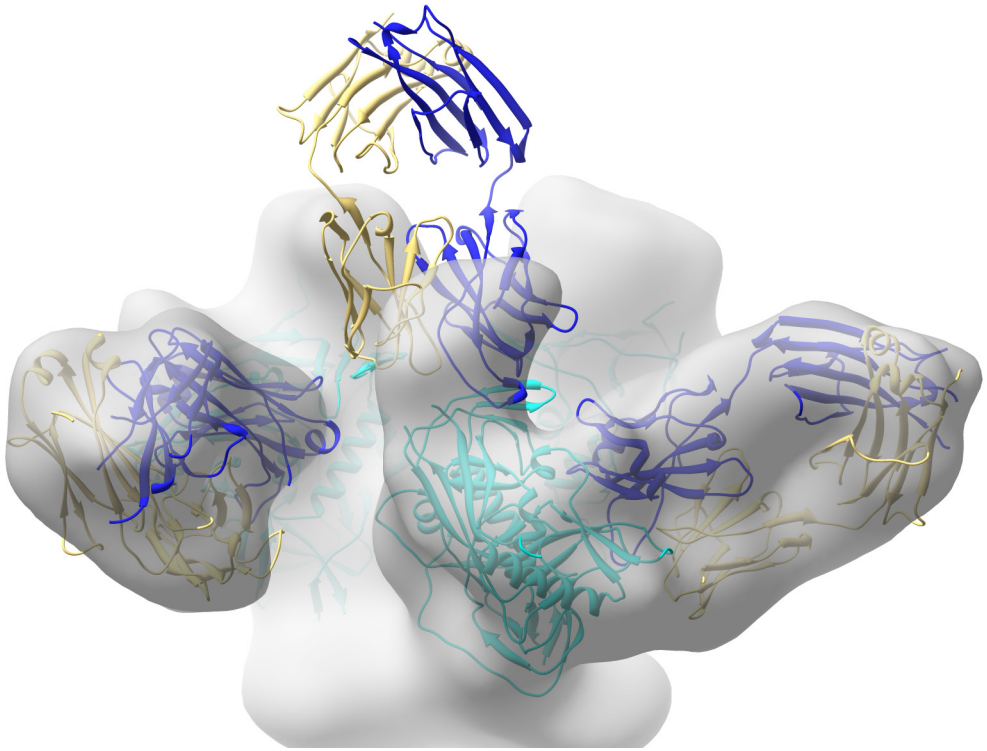

Figure S5. Triumvirate towers formed by VRC-PG04 inhibit CDi mAbs. Transparent surface representation of SOSIP.681 liganded to VRC PG04 image reconstructions with the appropriate crystal structures docked in and colored according to **Figure S4**. Superimposed onto the VRC PG04 liganded gp120core is the gp120core of the gp120 + sCD4 + CDi 17b ternary complex (PDB entry: 2NXY). Only the Fab region of the 17b antibody is shown. Steric clash between the 17b Fab fragment and V1/V2 inhibits 17b from binding the VRC PG04 liganded Env.