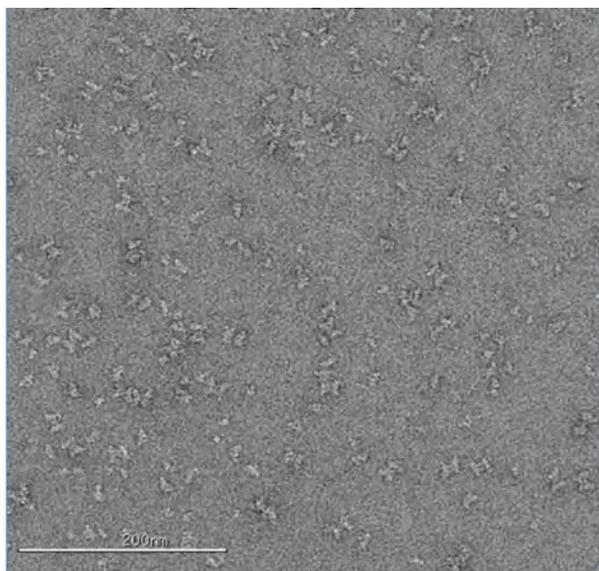


Fig. S1

a.



b.

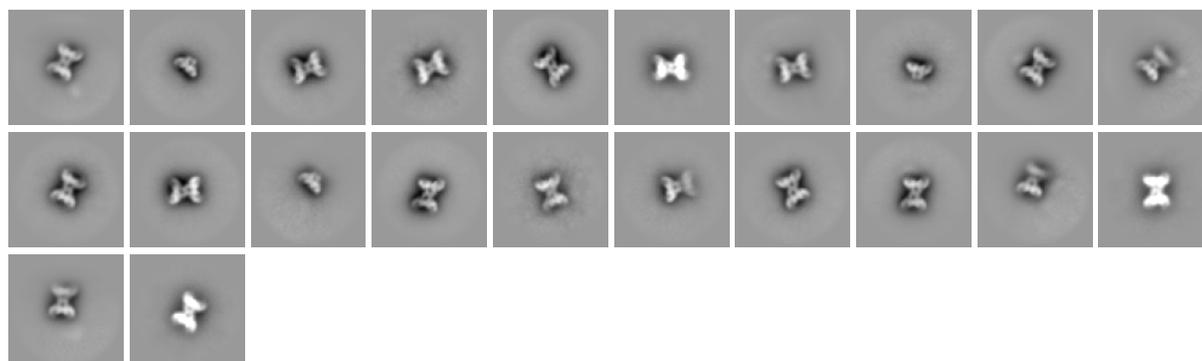


Fig. S2

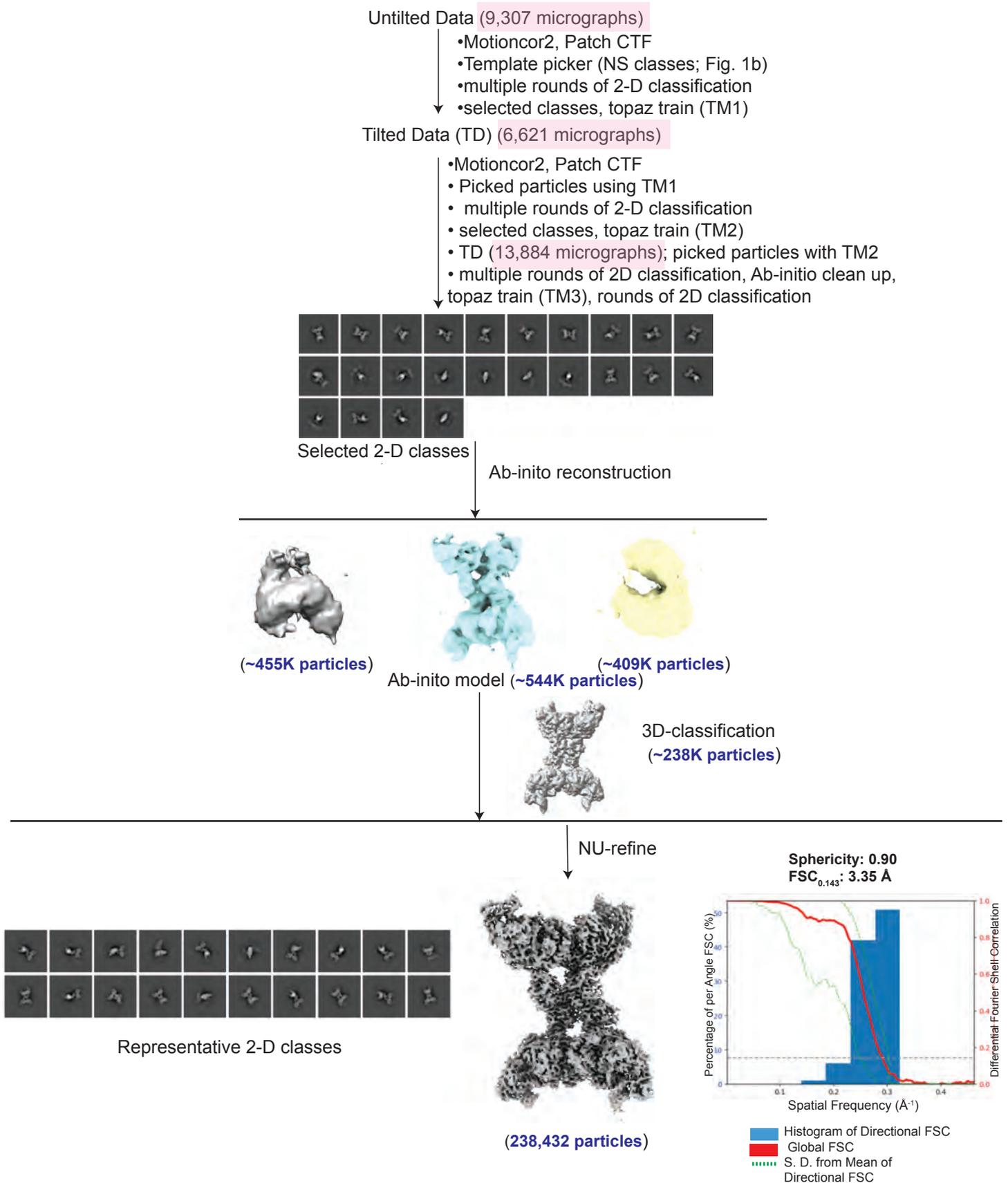
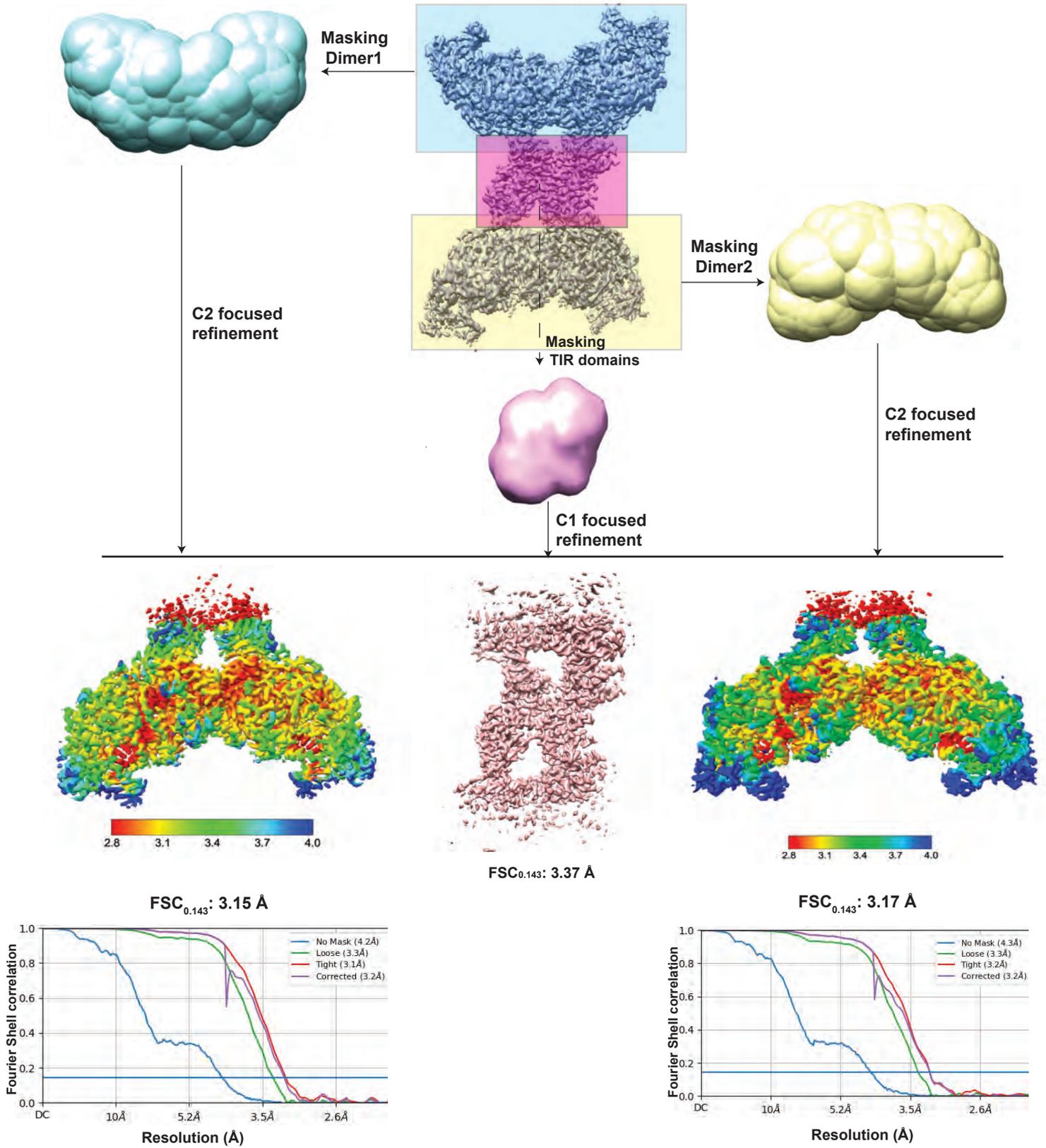
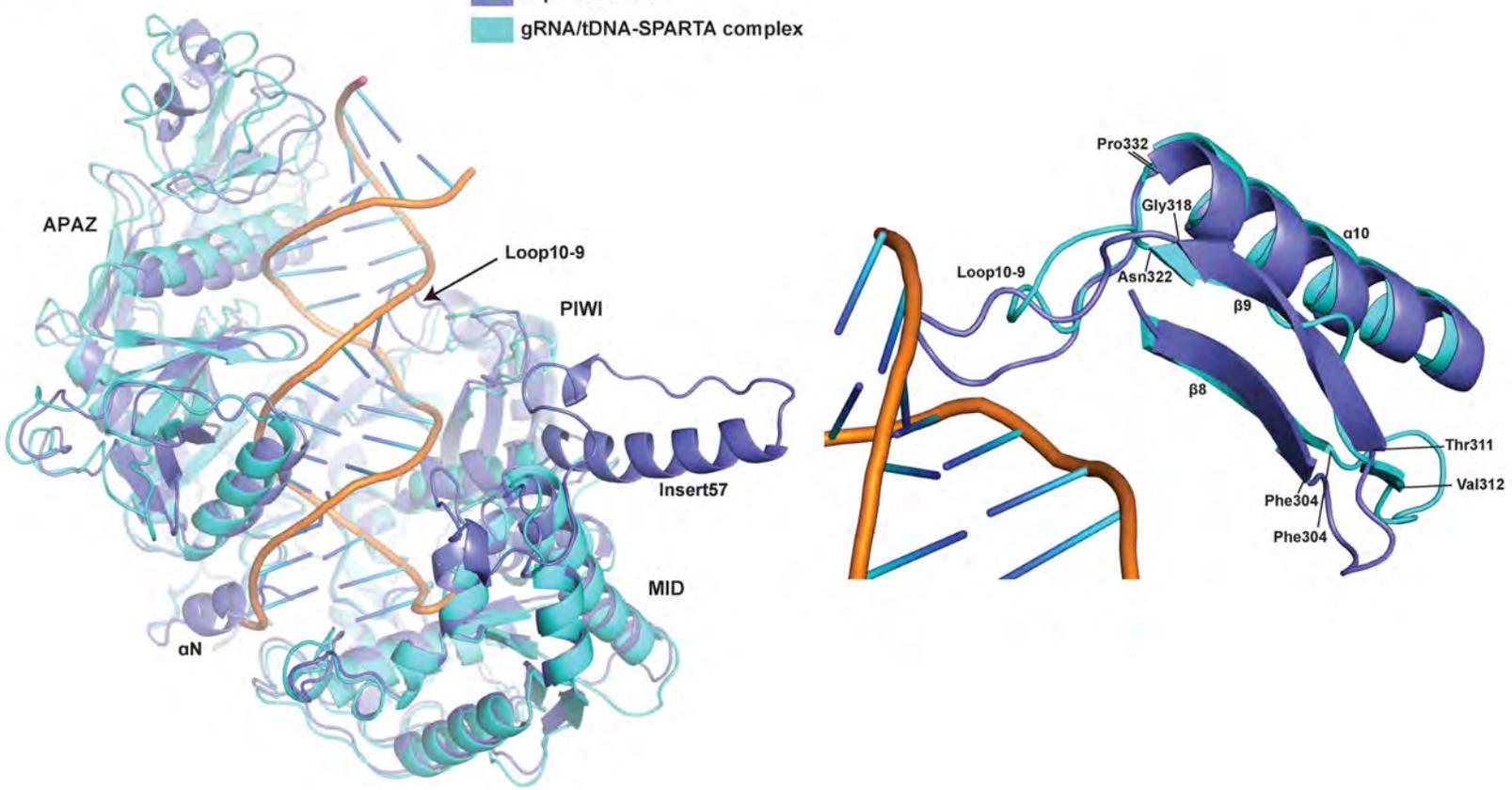


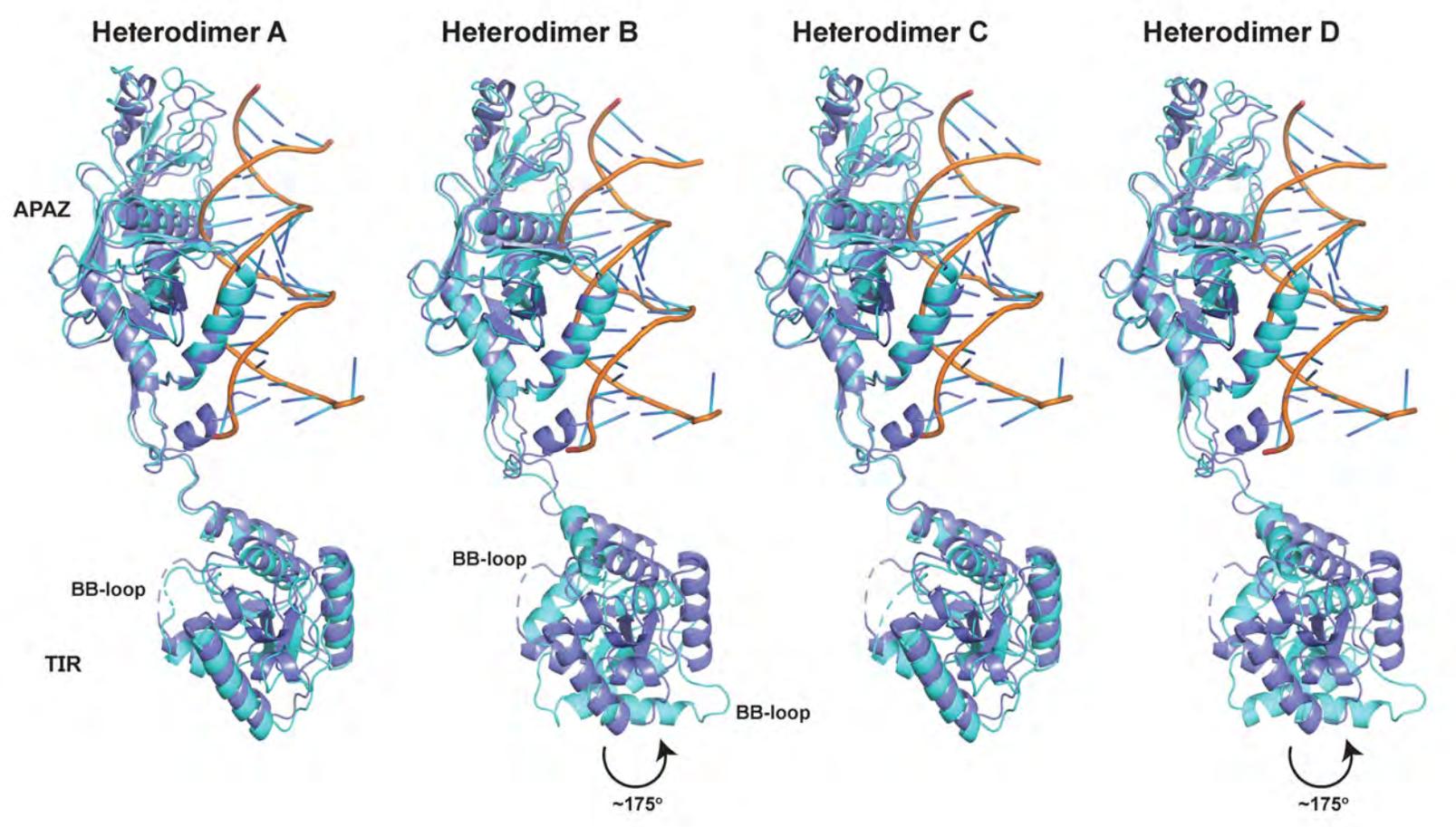
Fig. S3

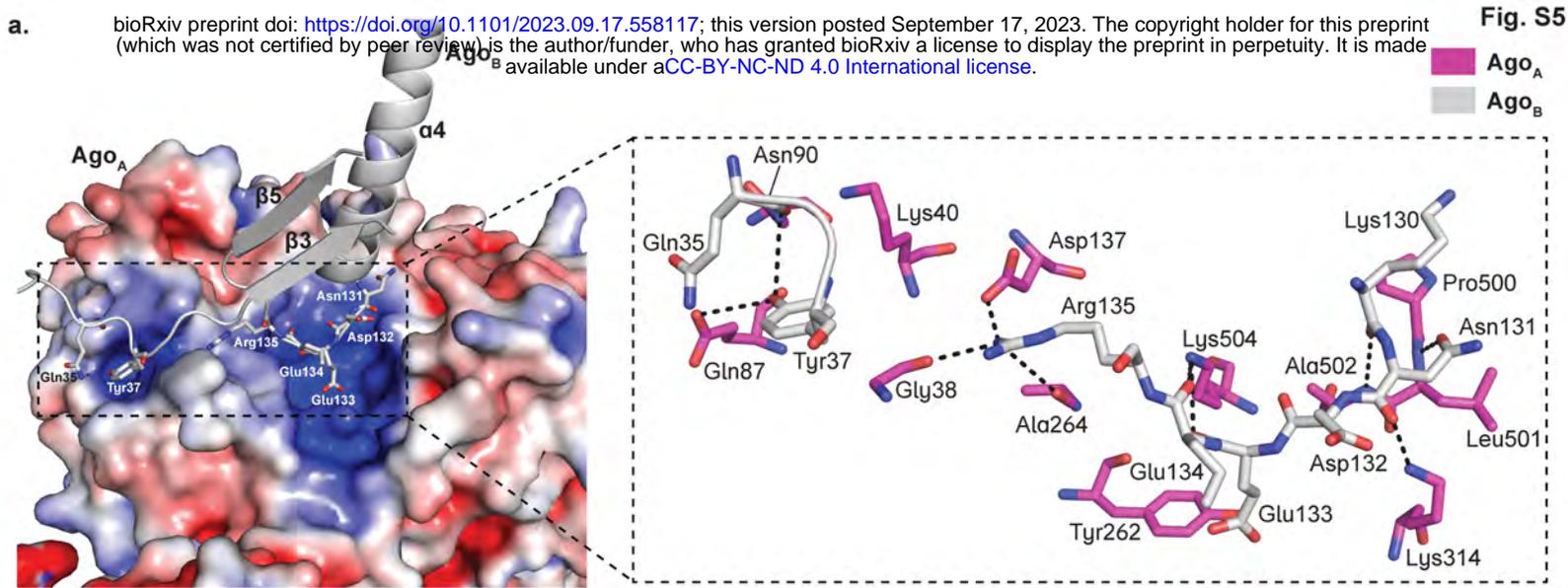


a.



c.



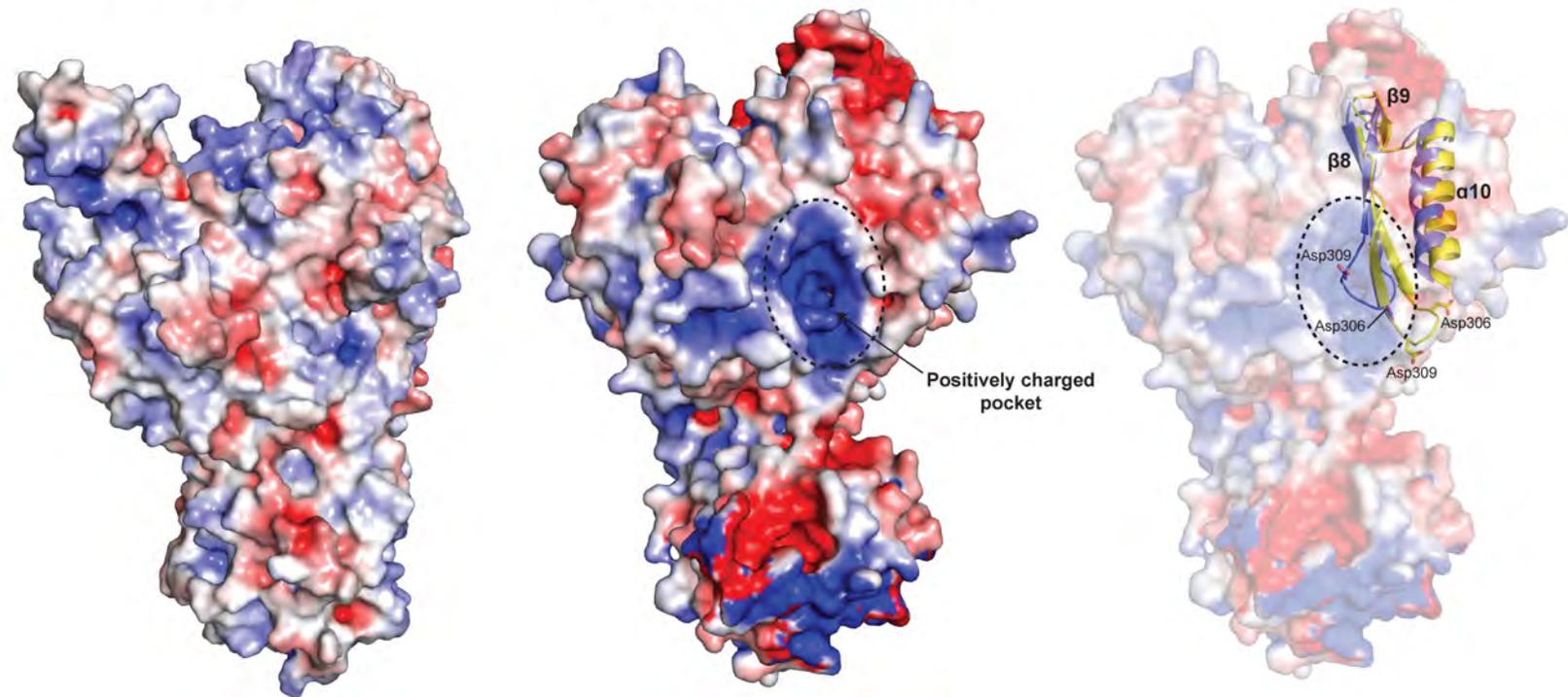


b.

Apo SPARTA
gRNA/tDNA-SPARTA complex

Apo SPARTA

gRNA/tDNA-SPARTA complex



Apo-SPARTA (PDB: 8U7B)	
Data collection	
Space group	P6 ₅ 22
Cell dimensions	
<i>a</i> , <i>b</i> , <i>c</i> (Å)	197.18 Å, 197.18 Å 183.4 Å,
α , β , γ (°)	90.00 90.00 120.00
Resolution (Å)	170.76-2.66 (2.99-2.66)*
<i>R</i> _{sym} or <i>R</i> _{merge}	14.8 (176.7)
<i>R</i> _{pim}	4.2 (55.4)
No. of unique reflections	40914
<i>I</i> / σ <i>I</i>	13.7 (1.7)
Completeness (%) (ellipsoidal)	96.2 (73.9)
Redundancy	13.3 (10.7)
CC(1/2)	1.00 (0.57)
Refinement	
Resolution (Å)	49.71-2.66
No. reflections	40895
<i>R</i> _{work} / <i>R</i> _{free}	18.6/24.0
No. atoms	
Macromolecules	7420
Ligands/Ions	2
Water	91
<i>B</i> -factors	
Macromolecules	69.0
Ligands/Ions	67.0
Water	63.9
R.m.s. deviations	
Bond lengths (Å)	0.003
Bond angles (°)	0.61
Ramachandran Plot	
Favored (%)	93.0
Allowed (%)	6.5
Outliers (%)	0.5

Table S2: CryoEM data collection, model refinement and validation statistics

SPARTA-gRNA-tDNA-Mg

PDB ID:8U72

EMDB IDs: EMD-41945, EMD-41947,

EMD-41948, EMD-41949, EMD-41966

Data collection and processing

Magnification (kV)	x81000
Voltage (kV)	300
Pixel Size (Å/pixel)	1.083
Electron dose (e ⁻ /Å ²)	51.11
Defocus range (µm)	0.8 - 2.5
Number of Micrographs	13,884
Tilt angle	30°, 45°
Number of particles	238,432
Nominal Map resolution (Å)	3.35
FSC threshold	0.143

Refinement (Phenix)

Model composition

Non-hydrogen atoms	29715
Protein residues	3469
DNA/other	148/4

R.M.S. deviations

Bond lengths (Å)	0.003
Bond Angles (°)	0.596

Validation

Molprobit score	1.90
Clashscore	7.07
Rotamer outliers (%)	0.33
Cβ outliers (%)	0.00

Ramachandran Statistics (%)

Favored	91.16
Allowed	8.78
Outliers	0.06