

Extended Data Table 1

	GajA (EMDB-36541) (PDB 8JQ9)	ATP-bound GajA (EMD-37915) (PDB 8WY4)	GajA/dsDNA (EMD-37916) (PDB 8WY5)	GajA/dsDNA local refinement (EMD-38058) (PDB 8X51)
Data collection and processing				
Magnification	105,000	105,000	105,000	
Voltage (kV)	300	300	300	
Electron exposure (e-/Å ²)	50	50	50	
Defocus range (µm)	-1.5 to -1.8	-1.5 to -1.8	-1.5 to -1.8	
Pixel size (Å)	0.84	0.84	0.84	
Symmetry imposed	D2	D2	D2	C1
Initial particle images (no.)	1,539,938	501,032	1,151,527	1,151,527
Final particle images (no.)	275,074	56,959	88,461	570,317
Map resolution (Å)	2.7	2.8	3.1	2.9
FSC threshold	0.143	0.143	0.143	0.143
Map resolution range (Å)	2.4 to 3.8	2.6 to 4.5	2.9 to 5.0	2.7 to 5.5
Refinement				
Initial model used (PDB code)	AlphaFold	8JQ9	8JQ9	8JQ9
Model resolution (Å)	2.9	3.1	3.5	3.1
FSC threshold	0.5	0.5	0.5	0.5
Model resolution range (Å)	50-2.9	50-3.1	50-3.5	50-3.1
Map sharpening <i>B</i> factor (Å ²)	-104.7	-80.9	-106.9	-87.3
Model composition				
Non-hydrogen atoms	14832	14864	16038	8101
Protein residues	1816	1804	1770	885
Nucleotide residues	0	0	76	42
Ligands	0	4	4	2
<i>B</i> factors (Å ²)				
Protein	48.19	43.86	39.80	44.24
Nucleotide			54.60	43.09
Ligand		117.89	30.56	23.64
R.m.s. deviations				
Bond lengths (Å)	0.004	0.007	0.008	0.007
Bond angles (°)	0.760	1.170	1.369	1.243
Validation				
MolProbity score	1.44	1.42	1.43	1.42
Clashscore	3.18	2.75	2.95	1.70
Poor rotamers (%)	0	0	0	0
Ramachandran plot				
Favored (%)	95.28	94.30	94.32	92.31
Allowed (%)	4.72	5.70	5.22	7.35
Disallowed (%)	0	0	0.46	0.34

	4:1 GajAB (EMDB-36569) (PDB 8JQC)	4:4 GajAB (EMDB-36563) (PDB 8JQB)	ATP/Mg ²⁺ -bound GajAB (EMDB-38071) (PDB 8X5N)	ATP/Mg ²⁺ -bound GajAB local refinement (EMDB-38070) (PDB 8X5I)
Data collection and processing				
Magnification	105,000	105,000	105,000	
Voltage (kV)	300	300	300	
Electron exposure (e-/Å ²)	50	50	50	
Defocus range (µm)	-1.5 to -1.8	-1.5 to -1.8	-1.5 to -1.8	
Pixel size (Å)	0.84	0.84	0.84	
Symmetry imposed	C1	D2	C1	C1
Initial particle images (no.)	360,360	632,358	504,335	504,335
Final particle images (no.)	78,983	21,115	190,783	41,197
Map resolution (Å)	3.4	3.2	2.8	3.0
FSC threshold	0.143	0.143	0.143	0.143
Map resolution range (Å)	3.3 to 6.8	3.2 to 6.5	2.8 to 6.2	3.0 to 7.2
Refinement				
Initial model used (PDB code)	8JQ9	8JQ9	8JQ9	8JQ9
Model resolution (Å)	3.8	3.3	3.0	3.2
FSC threshold	0.5	0.5	0.5	0.5
Model resolution range (Å)	50-3.8	50-3.3	50-3.0	50-3.2
Map sharpening <i>B</i> factor (Å ²)	-60.1	-59.9	-65.9	-50.0
Model composition				
Non-hydrogen atoms	18899	31664	23578	17130
Protein residues	2315	3876	2863	2063
Nucleotide residues	0	0	0	0
Ligands	0	0	12	12
<i>B</i> factors (Å ²)				
Protein	116.4	137.01	68.02	74.17
Nucleotide				
Ligand			109.60	132.13
R.m.s. deviations				
Bond lengths (Å)	0.004	0.006	0.028	0.009
Bond angles (°)	0.950	1.178	1.156	1.235
Validation				
MolProbity score	1.59	1.81	1.36	1.38
Clashscore	4.24	5.71	2.29	2.31
Poor rotamers (%)	0.24	0.31	0	0
Ramachandran plot				
Favored (%)	94.25	91.73	94.96	94.53
Allowed (%)	5.62	7.47	4.72	5.18
Disallowed (%)	0.13	0.81	0.32	0.29