Electronic Supplementary Material (ESI) for Physical Chemistry Chemical Physics.

Supporting information

Simulations of PKA RIα Homodimer Reveal cAMP-coupled Conformational Dynamics of Each Protomer and the Dimer Interface with Functional Implications

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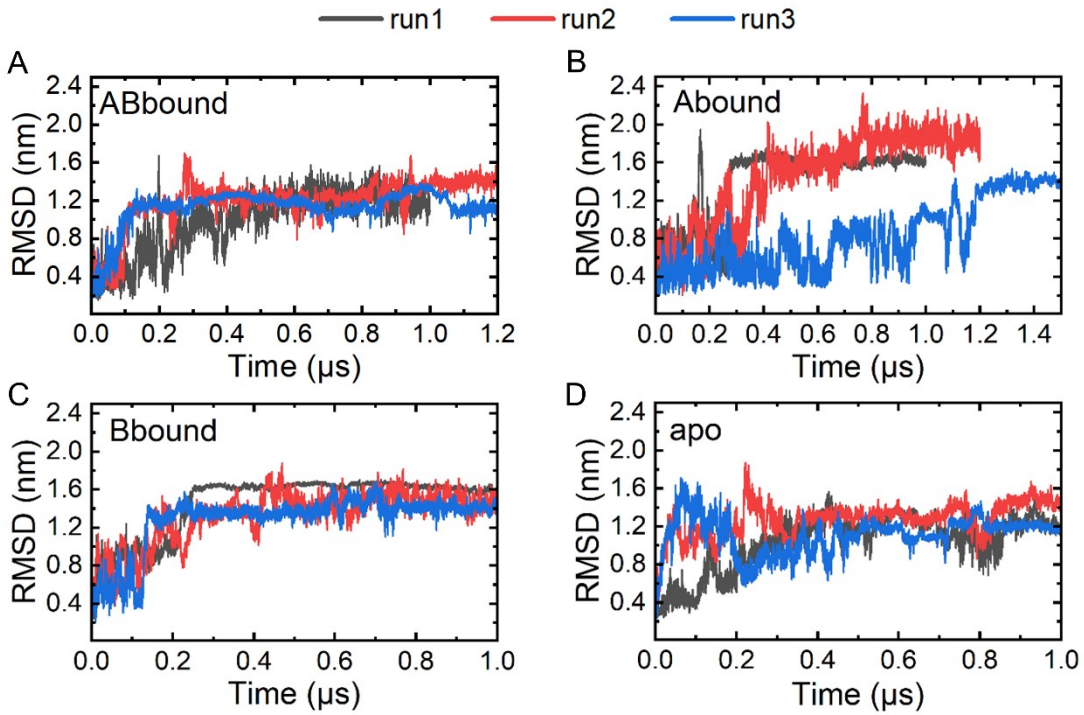

Figure S1. The overall RMSD of backbone atoms as reference to the starting structure as a function of time. For each system, results of three independent runs are shown in black, red and blue, respectively.

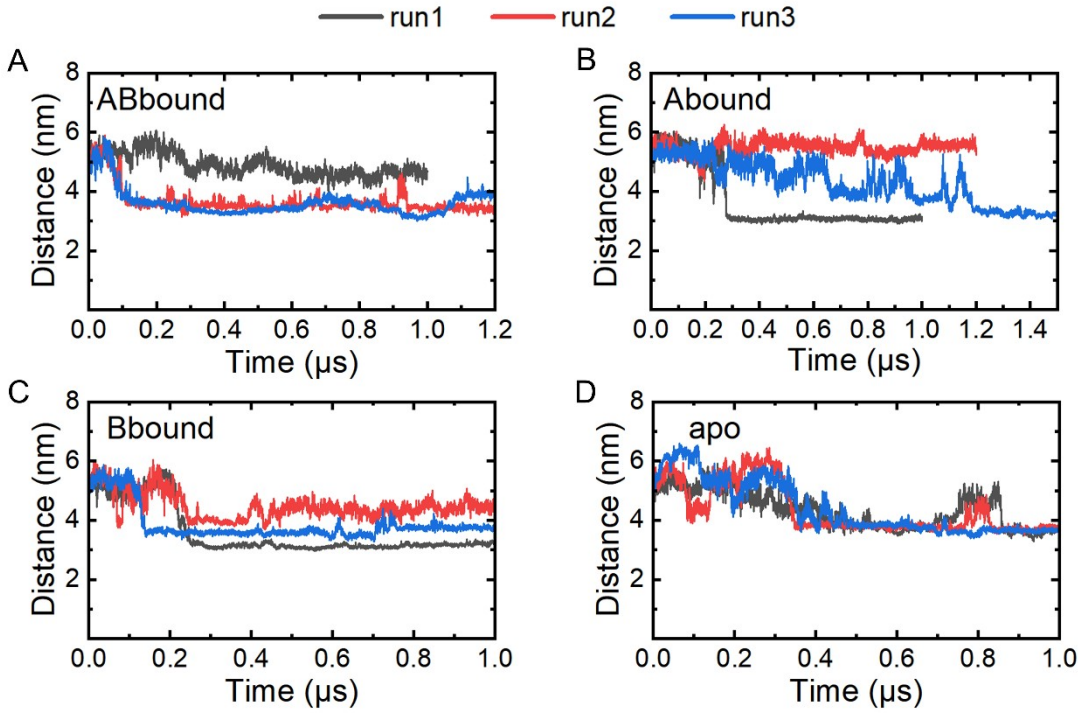

Figure S2. The center-of-mass distance between two R subunits as a function of time. For each system, results of three independent runs are shown in black, red and blue, respectively.

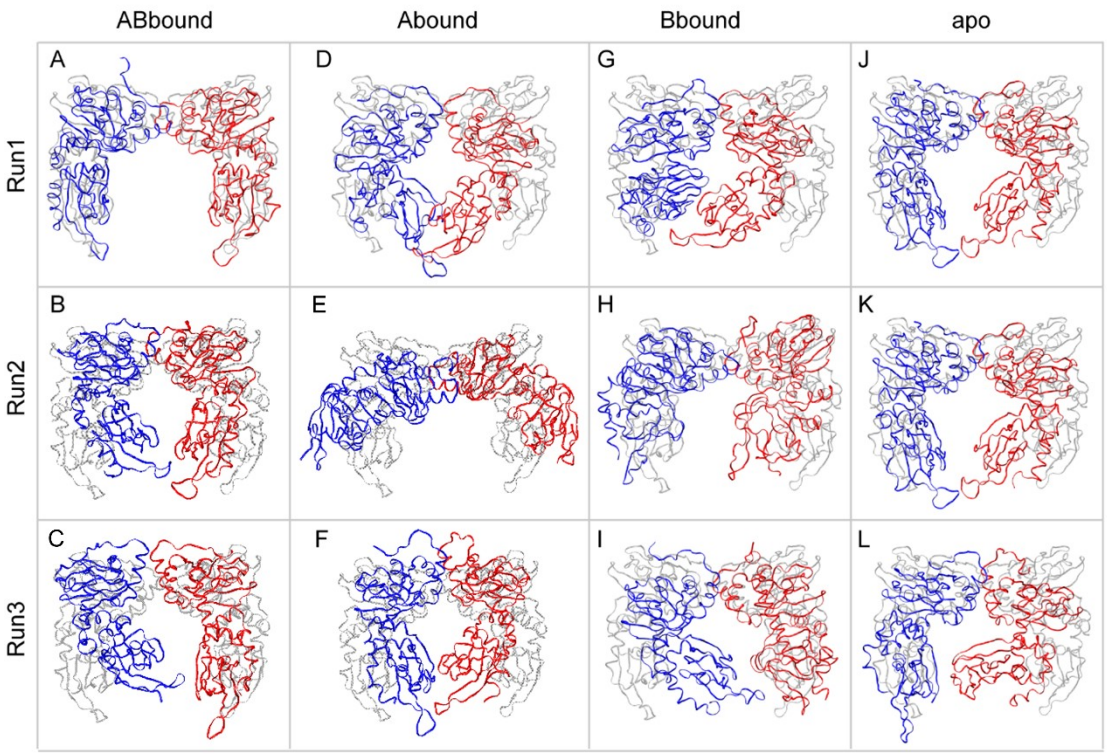

Figure S3. Comparison of the final homodimer structures in four systems to the crystal structure. Backbone superposition is over the N3A-N3A' interface. The crystal structure is in transparent gray. Two R subunits in the MD structures are in blue (chain A) and red (chain B).

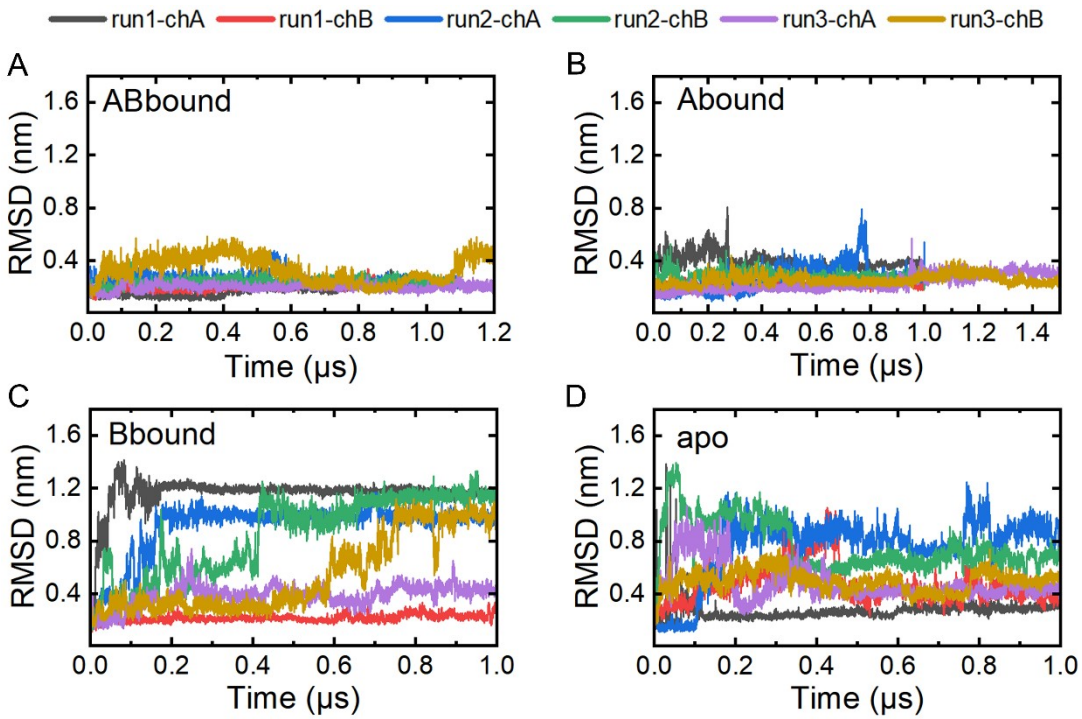

Figure S4. The backbone RMSD of each protomer as reference to the starting structure as a function of time. For each system, results of two protomers from three independent runs correspond to six curves and are shown in different colors.

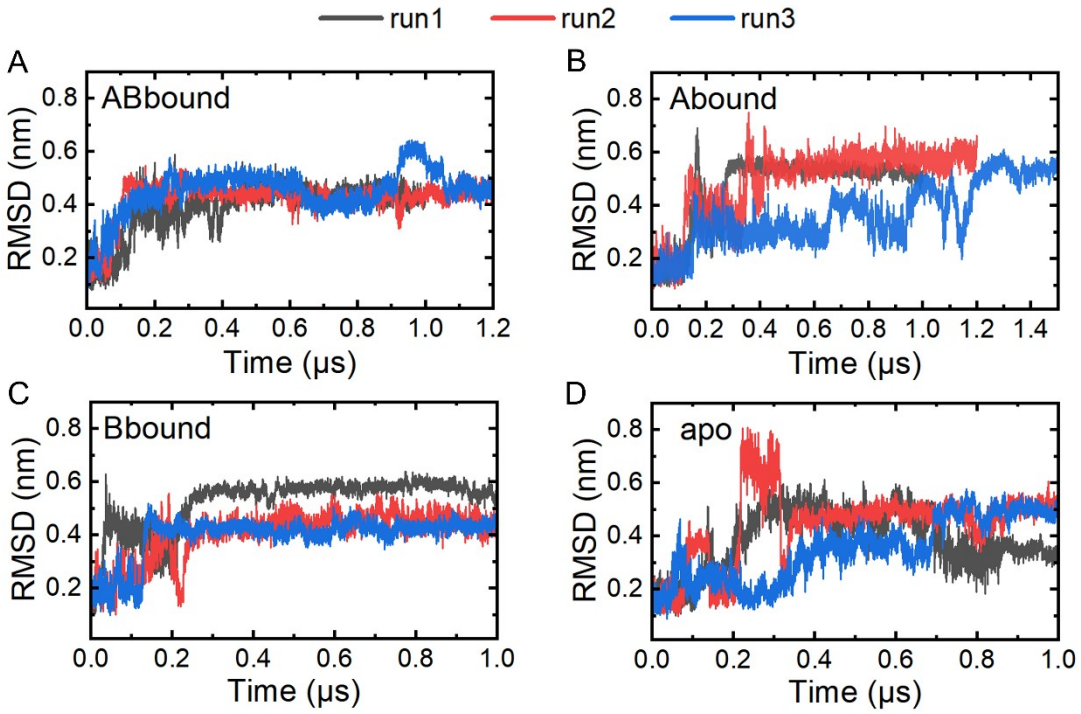

Figure S5. The backbone RMSD of the N3A-N3A' interface as reference to the counterpart in the starting structure as a function of time. For each system, results of three independent runs are shown in black, red and blue, respectively.

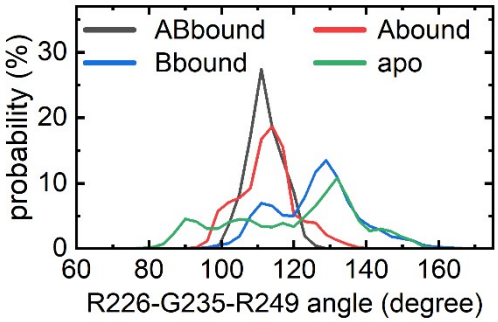

Figure S6. The probability distribution of the distortion angle of the B/C helix formed by the Cα atoms of R226, G235 and R249 in four research systems.

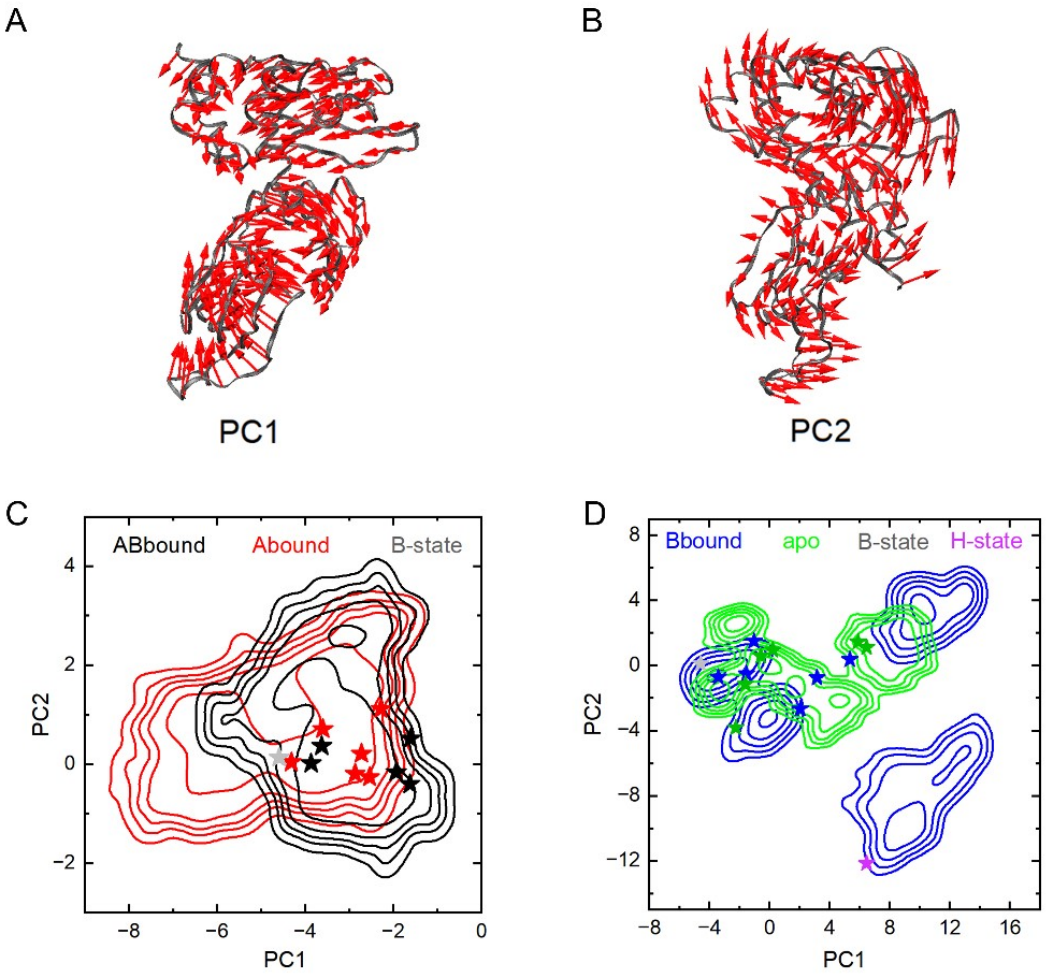

Figure S7. The motions corresponding to the first two principal components (PC1 and PC2) are illustrated in (A) and (B). Vectors starting from the Cα atoms indicate the direction and amplitude of atomic motions. To compare the conformations of individual R subunits in the monomeric and homodimeric states, we borrowed a series of representative conformations from previous simulations of monomeric R in the same four different cAMP-bound states and calculated their PC values. The corresponding positions on the FELs are labeled by colored stars (black for ABbound, red for Abound, blue for Bbound and green for apo) in (C) and (D). The positions of the B-state and H-state on the FELs are also labelled for reference.

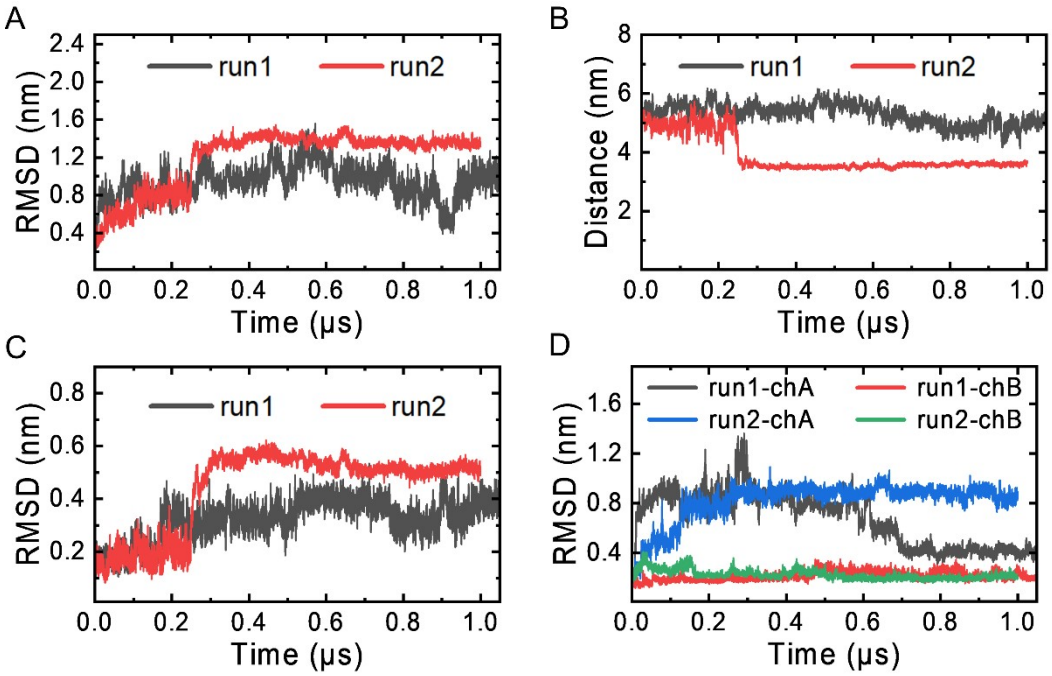

Figure S8. Convergence check of the 2B1A system is assessed by four physical quantities over time, including (A) the overall RMSD of backbone atoms, (B) the center of mass distance between two R subunits, (C) the backbone RMSD of the N3A-N3A' interface and (D) the backbone RMSD of each protomer. The starting structure is used as the reference in RMSD calculation.

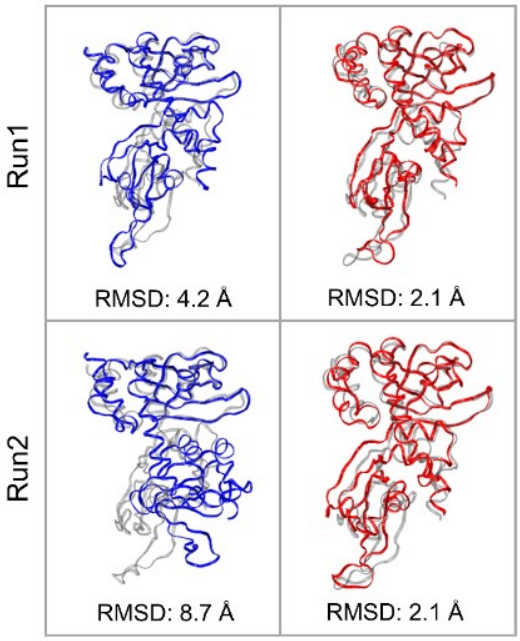

Figure S9. Final structures of two individual R subunits generated by two independent simulations of the 1A2B system (chain A in blue, chain B in red) are separately superimposed to the crystal structure (transparent gray). Backbone atoms of β-barrel:A are used for fitting.

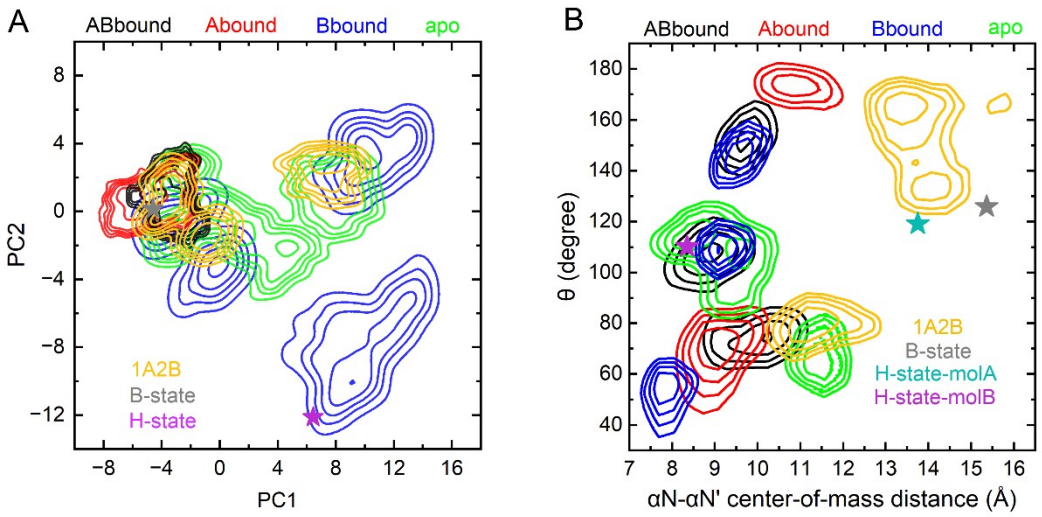

Figure S10. Free energy landscapes of the 1A2B system and comparison with the other four systems. (A) shows the free energy landscape of individual R subunits for 1A2B (orange contour line). Snapshots collected in the last 100 ns of two trajectories of the 1A2B system are projected onto the first two PCs obtained with the trajectories of ABbound, Abound, Bound and apo. Description of the other contour lines is the same as that in Figure 3. (B) shows the free energy landscape of the N3A-N3A' interface of the 1A2B system. Description of the *x* and *y* axis is the same as that in Figure 5.

Table S1. Backbone RMSD (Å) of the N3A-N3A' interface reference to the crystal structure of the homodimer of R subunit.

Run	ABbound	Abound	Bbound	Apo
Run1	4.3	5.3	5.8	3.5
Run2	4.5	5.9	4.4	5.2
Run3	4.6	5.3	4.4	5.1

Table S2. Average number of intermolecular contacts between four helices at the N3A-N3A′ interface in each simulation.

System	Run index	Number of contacts for different pairs of helices			
		αN-αN′	αN-αA′	αA-αN′	αA-αA′
ABbound	Run1	62.7	83.0	79.0	0.0
	Run2	72.2	68.3	88.2	0.0
	Run3	90.4	5.7	18.8	0.0
Abound	Run1	105.8	20.0	31.6	0.0
	Run2	90.0	**6.0**	**88.9**	0.3
	Run3	54.1	**15.0**	**147.0**	0.0
Bbound	Run1	89.5	19.2	33.6	0.0
	Run2	53.9	103.7	97.3	0.0
	Run3	37.6	110.3	104.1	0.0
apo	Run1	43.7	93.9	106.6	0.0
	Run2	76.7	89.1	97.9	0.0
	Run3	29.6	122.7	109.3	0.0
Crystal structures	B-state	20	77	94	0
	H-state molecule A	46	51	49	0
	H-state molecule B	180	0	0	0