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Supplemental Information

Cryo-EM Structures of the Hsp104 Protein Disaggregase Captured in the ATP Conformation

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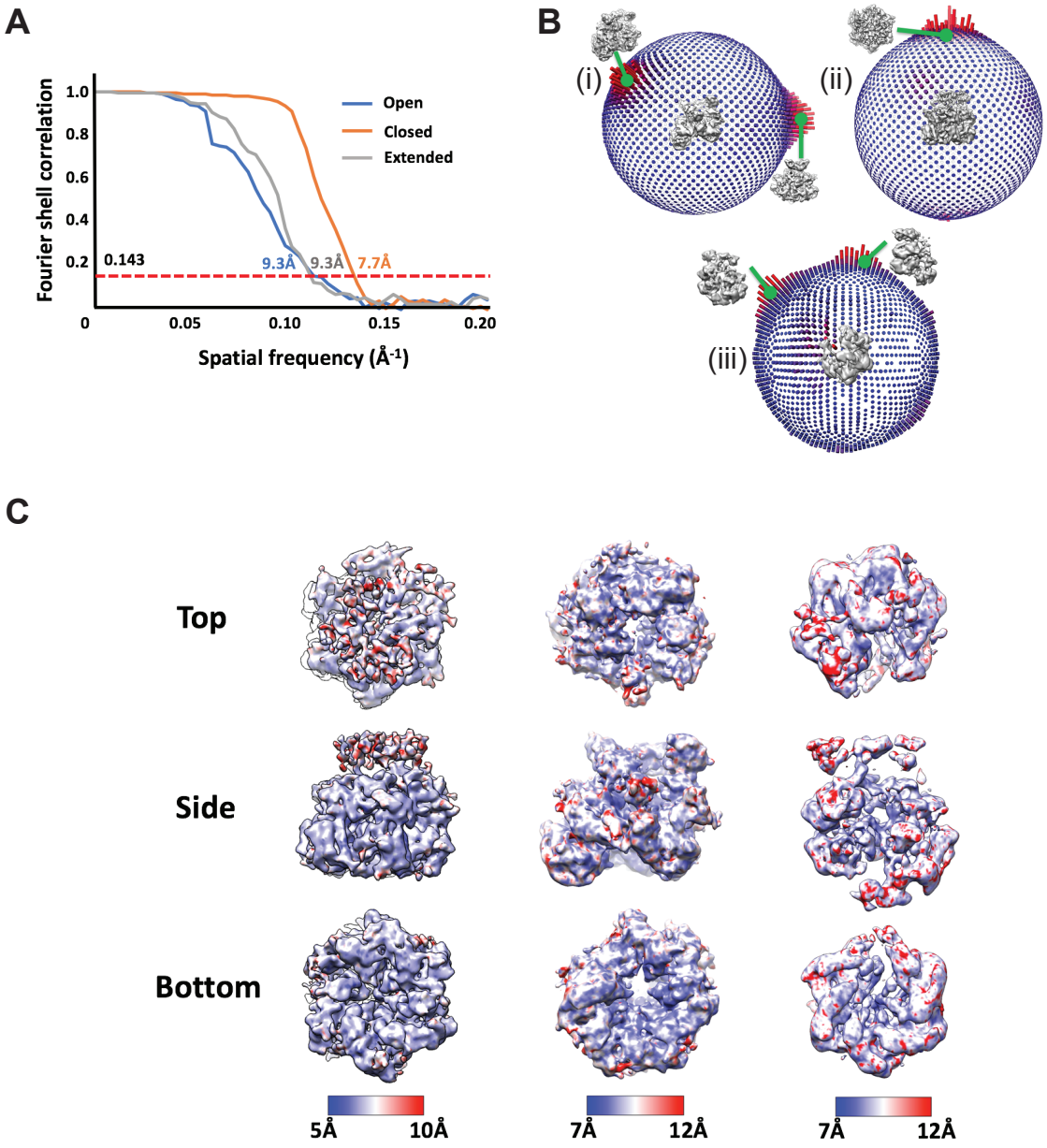

Figure S1. Three-dimensional reconstructions of $Hsp104_{DWB}$-ATP. Related to Figure 2.
(A) Gold-standard Fourier shell correlation (FSC) of $Hsp104_{DWB}$ in the open, closed, and extended conformation. **(B)** Angular distribution histogram of open (i), closed (ii), and extended (iii) conformation of $Hsp104_{DWB}$. **(C)** Local resolution of the reconstruction of the closed (left), open (middle), and extended conformation (right column) estimated by ResMap (Kucukelbir et al., 2014). Top, side, and bottom views are shown.

Table S1. Data Collection, Processing, and 3D Reconstruction. Related to Figure 2.

Data Collection and 3D Reconstruction	**Instrument/Software/Settings**	**Specifics**
Grid	Cu (Quantifoil)	400 mesh 1.2/1.3
Cryo-specimen freezing	Vitrobot (FEI)	Single blot, 95% relative humidity, 2 sec blot time
Electron microscope	Polara G2 (FEI)	300 keV
Detector	Gatan K2 Summit	4K X 4K, 5μm pixel size
Sampling interval	0.84 Å/Pixel	Super-resolution, 1.68 Å/Pixel (2X2 binned)
Exposure rate on specimen	5 $e^-/Å^2$/sec	5 frames/sec
Exposure time	7.6 sec	Total dose 38 $e^-/Å^2$
Drift correction	Unblur with exposure filter	1,394 movies
Defocus range	0.7 – 4 μm	
Defocus determination	CTFFIND4	
Particles picked	RELION 1.4	
Particle box size	160 X 160	
Number of particles used	Closed (56,859) Open (25,582) Extended (33,666)	out of 256,378 boxed particles
Initial map generation	EMAN 2.1	
Map refinement	RELION 1.4	
Resolution	7.7 Å for Closed 9.3 Å for Open 9.3 Å for Extended	Gold standard FSC at 0.143
Modeling	Chimera	