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Supporting information for article:

High-speed High-resolution Data Collection on a 200 keV Cryo-TEM

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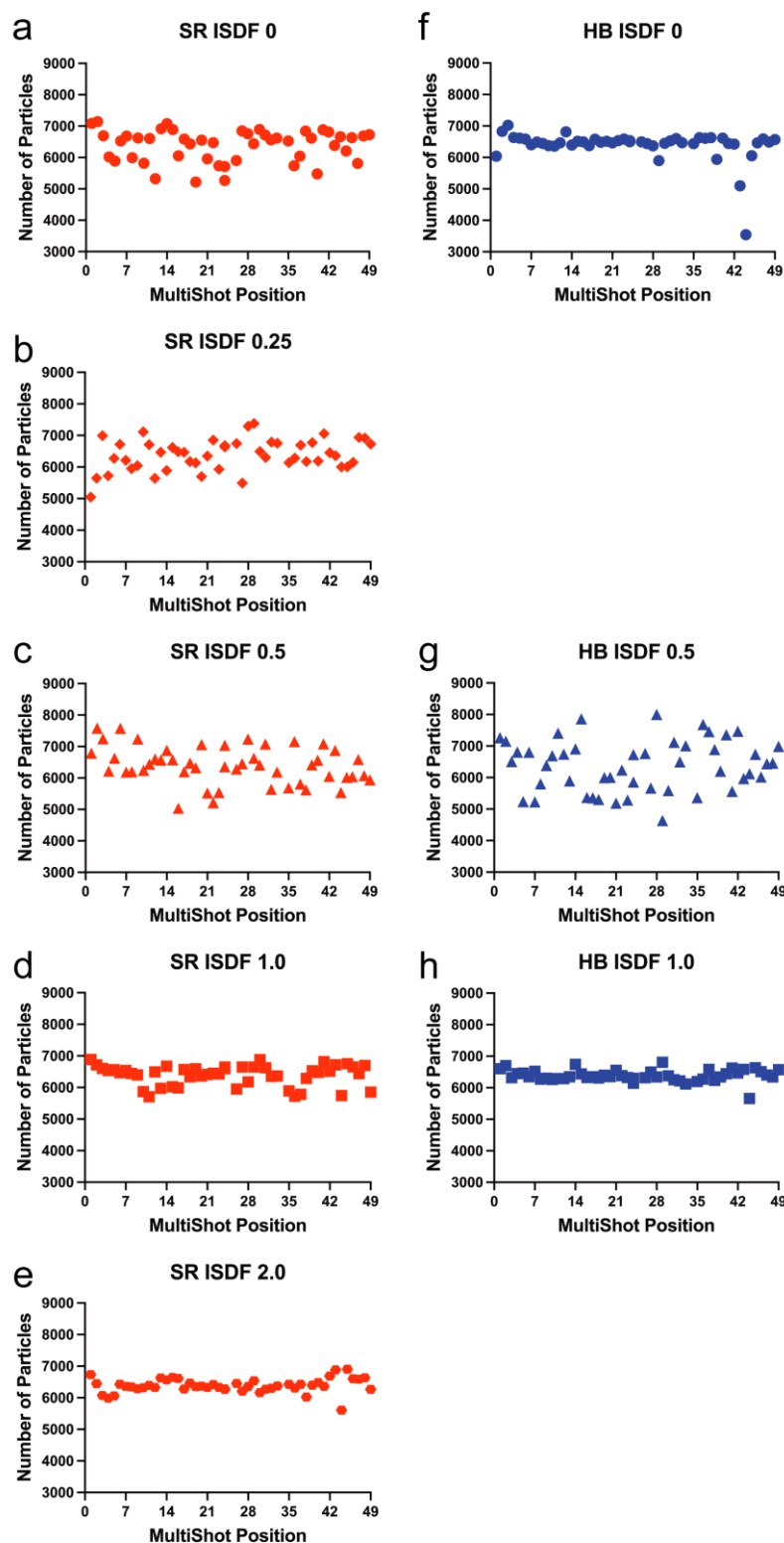


Figure S1. Number of Particles by Multishot Position for Data Collection with Different Image Shift Delay Factors First-level heading. (a-h) Number of particles contributed to final EM map according to order taken within the multishot array. Dataset collected in super-resolution (SR, indicated in red) or hardware binned (HB, indicated in blue). Image shift delay factor (ISDF) was set to either 0, 0.25, 0.5, 1 or 2.

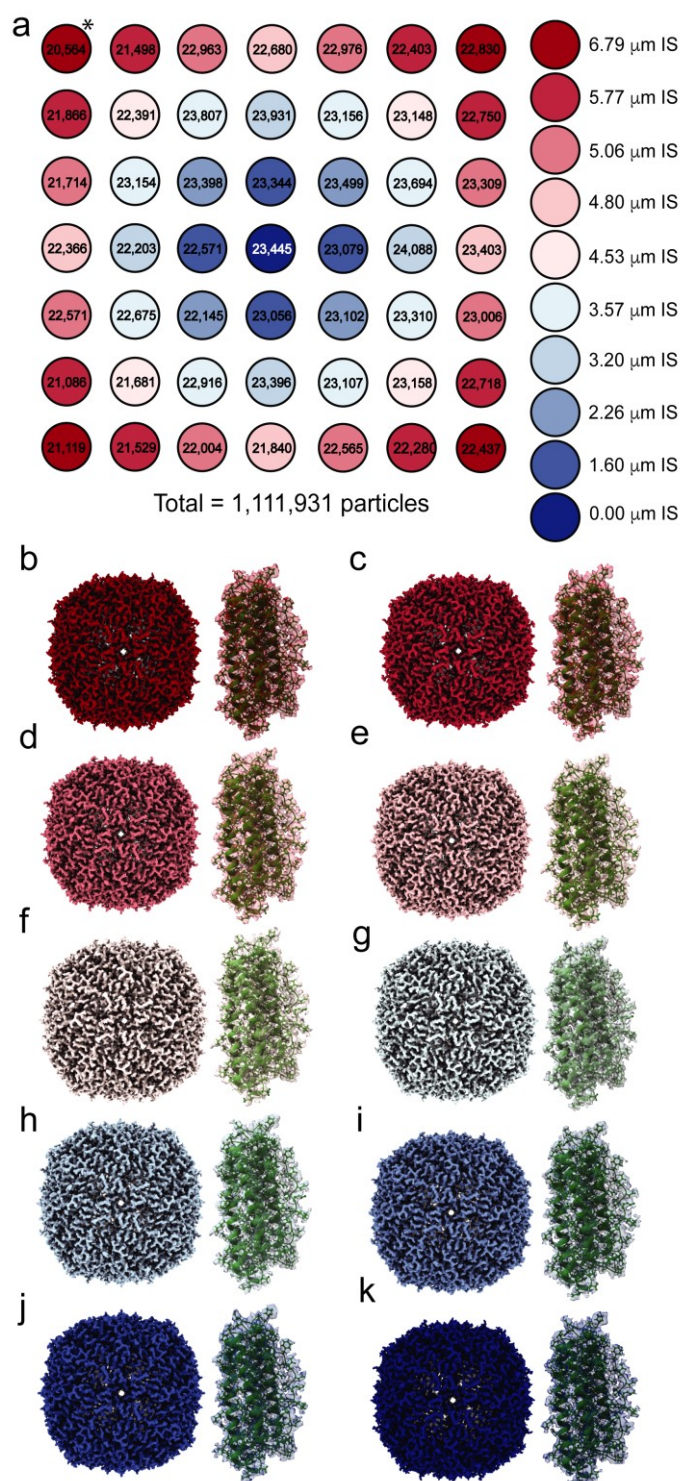


Figure S2. Particle Contribution to Final EM Map by Multishot Position. Particles (1,111,931) included in the final 1.78 Å EM map were separated into 10 groups based on image shift (IS) distance and processed independently. (a) The number of particles in each of the 7x7 multishot positions are indicated in the coloured circles. Position 1, marked here with an asterisk, was the only position that showed a significant difference, contributing 10% fewer particles than average. (b-k) EM maps reconstructed from particles grouped together based on image shift distance. The EM maps showed no significant difference in resolution or B-Factor.

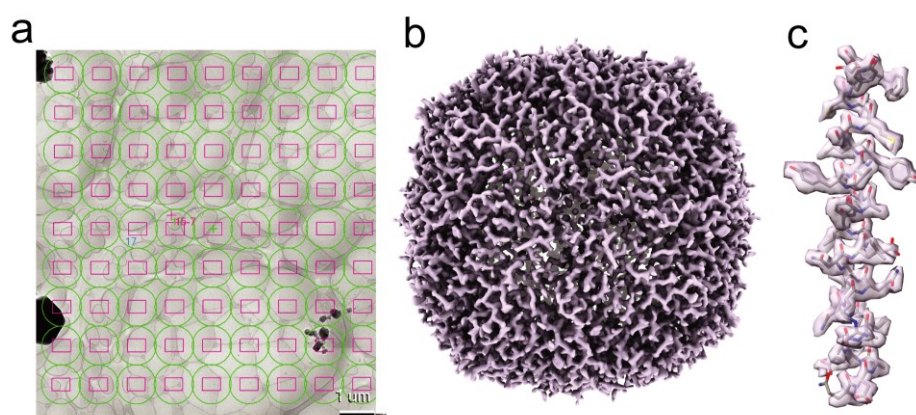


Figure S3. High-speed Data Collection on a Lacey TEM grid. (a) A single 9x9 multishot array on a Lacey carbon TEM grid. Green circles indicate beam diameter, the position of the Gatan K3 detector in pink. (b) Isosurface rendering of mouse apoferritin collected on a single grid square at 720 movies per hour. (c) Atomic model of mouse apoferritin PDB 6v21 docked into EM map, closeup of helix 1 GLN14 to ASP42.

Table S1

Electron Beam Diameter

50 μm Condenser				
Aperture				
<u>Spot Size</u>	<u>% C2 Parallel Illumination</u>	<u>Beam Diameter (μm)</u>	<u>Flux (e⁻/pixel/sec)</u>	<u>Dose Rate (e⁻/\AA^2/sec)</u>
1	46.461	1.73	29.94	36.16
2	44.829	1.599	25.54	30.84
3	43.2	1.67	15.58	18.81
4	41.891	1.632	10.47	12.64
5	40.826	1.695	6.16	7.44
6	39.288	1.567	3.84	4.64
7	38.468	1.64	1.82	2.2
8	37.952	1.62	0.6	0.72
Average Beam		1.64		
Diameter (μm)				

* Measurements made at nominal magnification of 54,900 X at the detector level, corresponding to pixel size of 0.88 \AA . The objective and selected area aperture were removed. Gun lens was set to 4.