Supplementary Information

Supplementary Tables

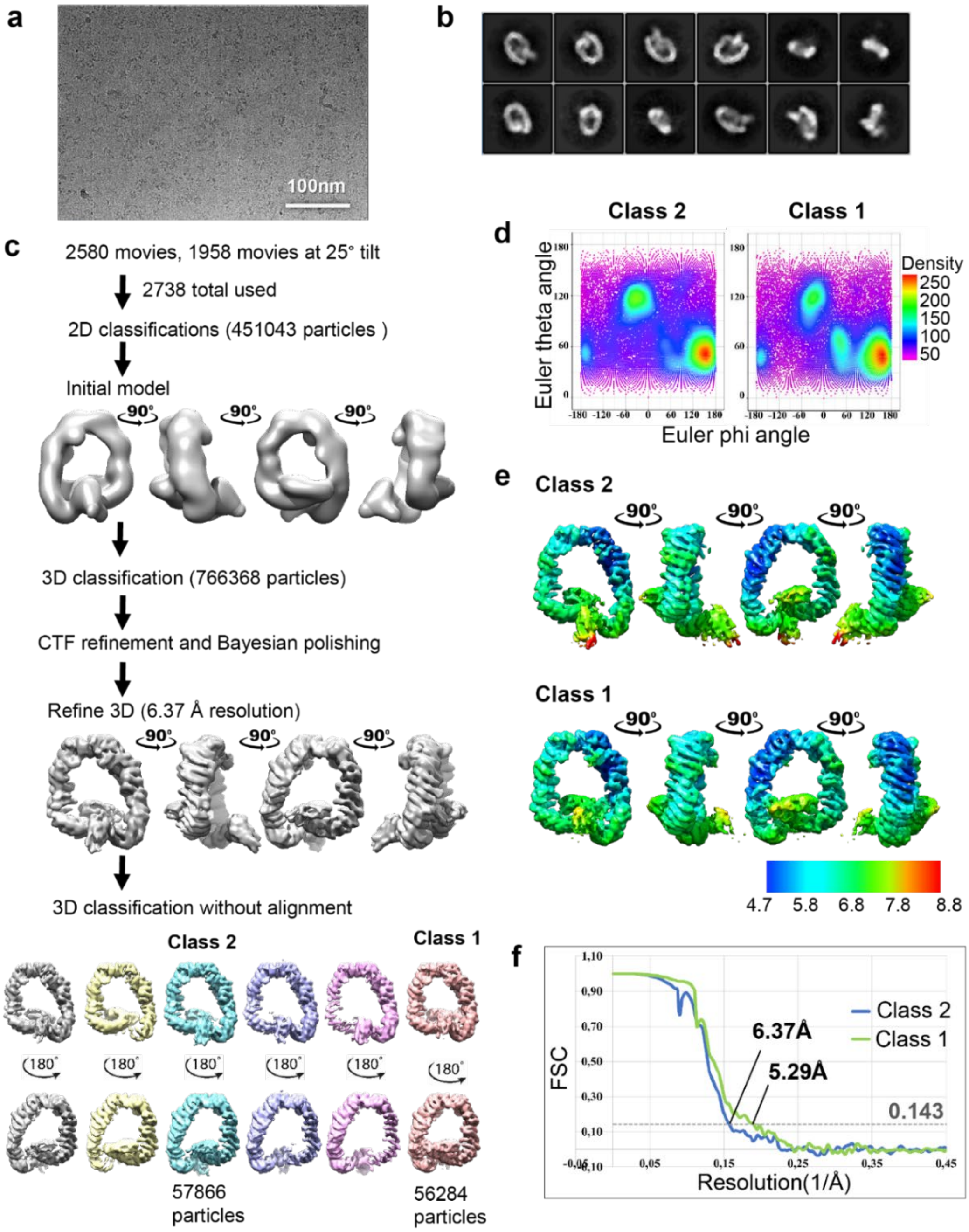
Supplementary Table 1 – Data collection and refinement statistics

	HUWE1$_N$ native	HUWE1$_N$ SeMet	HUWE1$_N$ TaBr
Data collection			
Space group	*P*1	*P*1	*P*1
Cell dimensions			
a, b, c (Å)	95.5, 96.2, 199.7	94.4, 95.3, 199.4	94.8, 96.2, 201.3
α, β, γ (°)	92.2, 100.5, 95.3	92.7, 100.2, 95.1	92.2, 99.6, 94.8
Resolution (Å)	196-3.05 (3.39-3.05)	98.0-3.42 (3.74-3.42)	95.8-4.99 (5.22-4.99)
R_{pim}	0.059 (0.358)	0.057 (0.573)	0.064 (0.485)
$I / \sigma I$	5.6 (1.6)	9.1 (1.5)	13.1 (1.7)
Ellipsoidal completeness (%)	89.4 (68.2)	84.9 (5.2)	91.6 (92.3)
Redundancy	3.8 (3.8)	10.7 (11.0)	13.9 (12.8)
Refinement			
Resolution (Å)	96-3.04		
No. reflections	75499 (3733)		
$R_{\text{work}} / R_{\text{free}}$ (%)	20.24/23.30		
No. atoms			
Protein	34901		
Ligand/ion	0		
Water	0		
B-factors (Å^2)			
Protein	104.8		
Ligand/ion	-		
Water	-		
R.m.s. deviations			
Bond lengths (Å)	0.006		
Bond angles (°)	0.85		

Supplementary Table 2 – Cryo-EM data collection, refinement and validation statistics

	Class 1 (EMDB-12318) (PDB 7NH1)	Class 2 (EMDB-12319) (PDB 7NH3)
Data collection and processing		
Magnification	105,000x	105,000x
Voltage (kV)	300	300
Electron exposure (e–/Å^2)	60.4	60.4
Defocus range (μm)	-2 to -3.5	-2 to -3.5
Pixel size (Å)	0.86	0.86
Symmetry imposed	C1	C1
Initial particle images (no.)	766368	766368
Final particle images (no.)	56284	57866
Map resolution (Å)	5.29	6.37
FSC threshold	0.143	0.143
Map resolution range (Å)	4.7-8.8	4.7-8.8
Refinement		
Initial model used (PDB code)	7BII	7BII
Model resolution (Å)	7.4	8.1
FSC threshold	0.5	0.5
Model resolution range (Å)		
Map sharpening *B* factor (Å^2)	-137	-178
Model composition		
Non-hydrogen atoms	17417	17417
Protein residues	2138	2138
Ligands	0	0
B factors (Å^2)		
Protein	488	563
Ligand	-	-
R.m.s. deviations		
Bond lengths (Å)	0.003	0.003
Bond angles (°)	0.639	0.654
Validation		
MolProbity score	1.99	2.02
Clashscore	15.58	16.86
Poor rotamers (%)	0	0
Ramachandran plot		
Favored (%)	95.7	95.8
Allowed (%)	4.23	4.24
Disallowed (%)	0.05	0

Supplementary Figures



Supplementary Figure 1 – CryoEM data processing workflow. (**a**) Representative summed micrograph from 2738 total used micrographs. (**b**) 2D classification results showing different views (**c**) Processing workflow. (**d-f**) The Euler angle distribution (**d**), local resolution maps (**e**), and Fourier shell correlation curves (**f**) for Class 1 and Class 2.