a

b

2,569 micrograph stacks → Motion correction / Manually check → 2,481 micrographs

2,481 micrographs → Autopick → 1,391,221 particles

1,391,221 particles → 2D classification → 583,408 good particles

583,408 good particles → 3D classification

(19%)

3D classification → 103,110 particles

103,110 particles → 3D classification

(26%) (23%) (16%) (25%) (6%) (4%)

3D classification

(39%)

3D refinement
Post processing

(4.3 Å)

EQ_tall

3D classification

(42%)

3D refinement
Post processing

(3.7 Å)

EQ_close

Supplementary information, Fig. S7 Image processing for the cryo-EM data of ATP-bound MlaF$_{EQ}$EDB in nanodiscs. a, Representative cryo-EM image and 2D class averages of cryo-EM particle images of ATP-bound MlaF$_{EQ}$EDB in nanodiscs. **b,** Flow chart for cryo-EM data processing. For details, see 'Data processing' in the Methods section.