



Fig. S10 Structure determination of ALMT1_{M60A/Al/pH5}.

a, Size-exclusion chromatography of AtALMT1 M60A mutant in 1 mM AlCl₃ at pH 5 on Superose 6 (GE Healthcare), and SDS-PAGE image of the final protein sample. **b**, Representative cryo-EM micrograph of ALMT1_{M60A/Al/pH5}. **c**, Flowchart of image processing for ALMT1_{M60A/Al/pH5} particles. **d**, The density map of ALMT1_{M60A/Al/pH5} colored by local resolution in Å. **e**, The Gold standard Fourier Shell Correlation (FSC) curve of the final 3D reconstruction of ALMT1_{M60A/Al/pH5}, and the FSC curve for cross-validation between the map and the model of ALMT1_{M60A/Al/pH5}. **f**, Sample maps at the 13 helices from ALMT1_{M60A/Al/pH5} at the contour level of 4 σ . **g**, EM maps of ALMT1_{Al/pH5} at the TM1–2 loop and TM5–6 loop regions at the contour level of 4 σ . The ribbon model is from the ALMT1_{M60A/Al/pH5} structure. **h**, EM maps of ALMT1_{M60A/Al/pH5} at the TM1–2 loop and TM5–6 loop regions at the contour level of 4 σ . The ribbon model is from the

- 1 ALMT1_{M60A/AI/pH5} structure. **i**, EM maps of ALMT1_{apo/pH5} at the TM1–2 loop and TM5–6 loop
- 2 regions at the contour level of 4σ . The ribbon model is from the ALMT1_{apo/pH5} structure.
- 3