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2 **Fig. S3 Structure determination of ALMT1_{apo/pH5}.**

3 **a**, Size-exclusion chromatography of the apo AtALMT1 at pH 5 on Superose 6 (GE Healthcare),
 4 and SDS-PAGE image of the final protein sample. **b**, Representative cryo-EM micrograph of
 5 ALMT1_{apo/pH5}. **c**, Flowchart of image processing for ALMT1_{apo/pH5} particles. **d**, The density map
 6 of ALMT1_{apo/pH5} colored by local resolution in Å. **e**, The Gold standard Fourier Shell Correlation
 7 (FSC) curve of the final 3D reconstruction of ALMT1_{apo/pH5}, and the FSC curve for cross-
 8 validation between the map and the model of ALMT1_{apo/pH5}. **f**, Sample maps at the 13 helices from
 9 ALMT1_{apo/pH5} at the contour level of 4σ.

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