

Figure 1 hTRPV1 purification and apo cryo-EM structure determination. **A** Size exclusion chromatograph where fractions used for cryo-EM are highlighted in grey. **B** SDS-PAGE showing single band for hTRPV1. **C** Apo hTRPV1 representative cryo-EM micrograph. **D** The three most populated reference-free 2D class averages from the cryo-EM dataset collected at ASU. **E** FSC curve with 3.04 Å resolution displayed at top of plot, no mask resolution shown in blue, loose mask shown in green, tight mask shown in red and corrected shown in purple. **F** Apo hTRPV1 3D reconstruction in grey and corresponding model in red. The side view is displayed on the left and top view shown on the right.

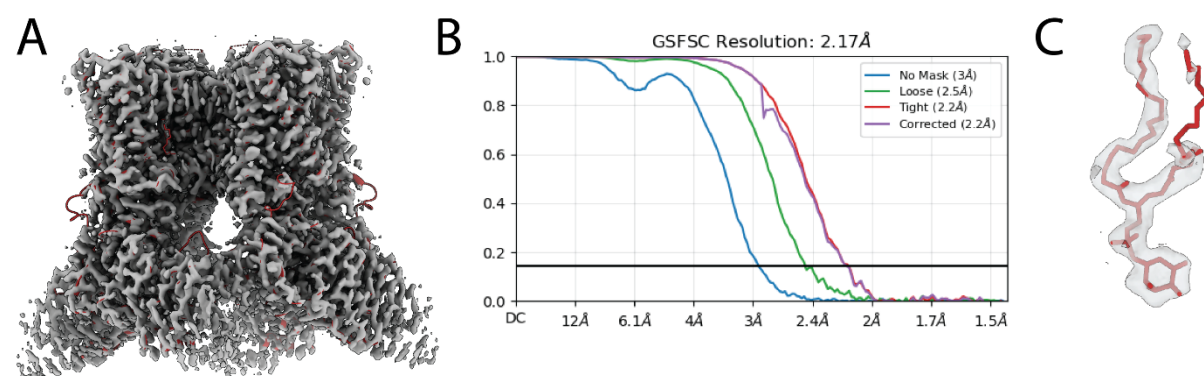


Figure 2 Agonist-bound hTRPV1 cryo-EM structure **A** 3D reconstruction for hTRPV1 with agonist bound side view in grey and model in red. **B** FSC curve with 2.17 Å resolution displayed at top of plot, no mask resolution shown in blue, loose mask shown in green, tight mask shown in red and corrected shown in purple. **C** Endogenous phosphatidylinositol lipid EM density displayed in transparent grey and the modeled in lipid in red.

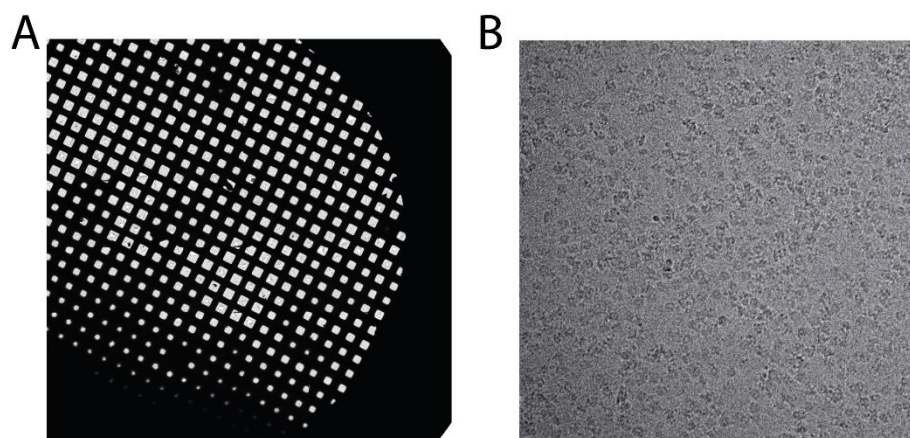


Figure 3 hTRPV1 bound to Mavatrip grid screening. **A** Atlas image for entire grid with hTRPV1 bound to Mavatrip. **B** Micrograph for optimal ice condition shows monodisperse particles with good particle spread.