

Fig. S3: Cryo-EM data analysis of the active NDP-α-MSH-MC4R-Gs-Nb35 complex.

- (a) Representative cryo-electron micrograph. The scale bar corresponds to 50 nm in the image.
- (b) Distribution of projection directions as estimated during homogeneous refinement with program cryoSPARC.
- (c) Global resolution estimation by Fourier shell correlation calculations (FSC = 0.143 cutoff) after "gold standard" refinement. The light blue curve was calculated without masking, the black curve by applying a spherical mask and the red one after phase randomization using a soft-mask.
- (d) Representative 2D class averages confirm random distribution of projection directions. The scale bar corresponds to 10 nm in the image.
- (e) Representation of local resolution estimation determined with cryoSPARC. The final cryo-EM density is colored according to the local resolution ranging from dark blue (2.4 Å) to red (4.4 Å).