



Figure S3. Overall maps of 30S under 2.5 and 10 mM Mg²⁺ condition. **a, b.** The 30S peaks of the sucrose gradients for BL21 DE3 at 2.5 mM Mg²⁺ (**a**) or 10 mM Mg²⁺ (**b**) were analyzed by cryoEM. Helix h16/h17, h18, h27, h44 and S12 were colored in yellow, blue, green, magenta and red, respectively. **c, d.** The Fourier Shell Correlation (FSC) curve for 2.5 mM (**c**)

and 10 mM (**d**) Mg^{2+} were calculated in Relion 3.1.0 and gold standard criteria with a value of 0.143 was used. **e, f**. The sharpened density map colored by local resolution in surface and slice views for the 30S under 2.5 (**e**) and 10 mM (**f**) Mg^{2+} concentration.