

Figure S5. Manual molecular replacement. Automated molecular replacement did not accurately place the vault 2-fold at a crystal 2-fold (compare Supporting Figs. S4, S6). The vault cryo-EM electron density is shown in cross-section. **(a)** The half-vault density was scaled smaller by 0.96. The center point of the whole vault was translated to **(0,0,0)** (black dot), placing the high-symmetry vault axis on the orthogonal **Z** axis. **(b)** The density was rotated -13.7° around the **Y** axis to position the density in the crystal coordinate system. (The crystal β angle is 123.8° between the **X** and **Z** axes.) The crystal 2-fold along **Y** (perpendicular to the black dot at **(0,0,0)**) generated the whole vault (see Fig. S6). The density rotation program thinned the vault shell. Thickness and isotropic shrinkage of the vault were adjusted to pack the cell (see *Methods* and Text S7). The two figure components were made at the same scale with CCP4 program MAPSLICER [1], then combined and labeled with Photoshop.

1. CCP4 (1994) The CCP4 suite: programs for protein crystallography. *Acta Crystallogr D Biol Crystallogr* 50: 760-763.

