Supplementary Information

Four levels of hierarchical organization including non-covalent chainmail brace the mature tumor herpesvirus capsid against pressurization

Z. Hong Zhou, Wong Hoi Hui, Sanket Shah, Jonathan Jih, Christine M. O'Connor, Michael B. Sherman, Dean H. Kedes, Stan Schein

Inventory of Supplementary Information

Four supplementary figures (Fig. S1 related to Fig. 1; Fig. S2 related to Fig. 3; Fig. S3 related to Fig. 5; Fig. S4 related to Figs. 2d and 6g)

Eight supplementary movies (Movie S1 related to Fig. 1b; Movie S2 related to Fig. 1d; Movie S3 related to Fig. 2; Movie S4 related to Fig. 3; Movie S5 related to Fig. 4; Movie S6 related to Fig. 5; Movie S7 related to Fig. 6; Movie S8 related to Fig. 7)

Supplementary Movie Legends:

Movie S1, related to Fig. 1b. Radially color-coded RRV capsid reconstruction.

Movie S2, related to Fig. 1d. Structural components of RRV capsid.

Movie S3, related to Fig. 2. Chainmail.

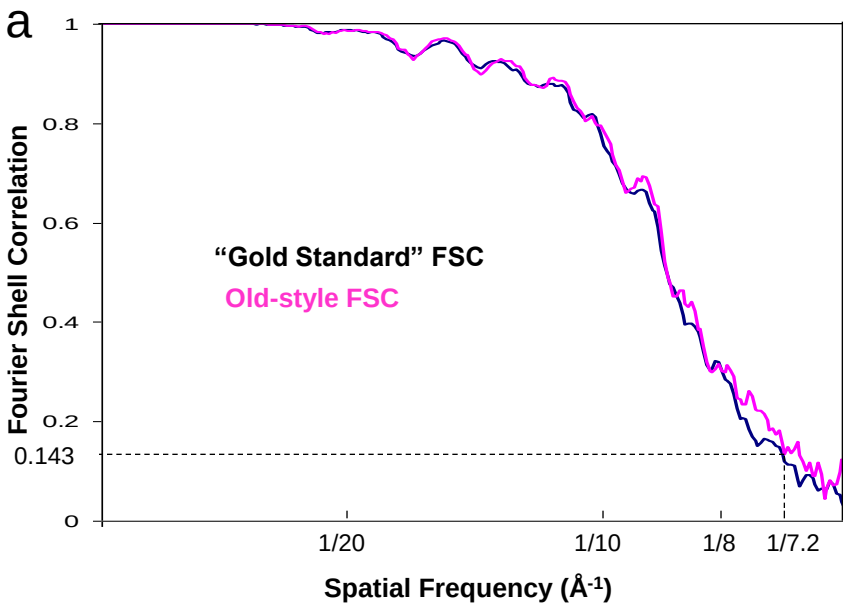
Movie S4, related to Fig. 3. Hexon subunit.

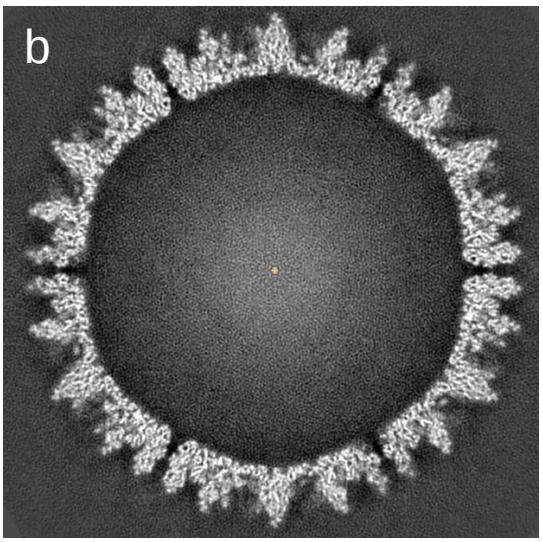
Movie S5, related to Fig. 4. MCP-MCP interaction sites in hexon.

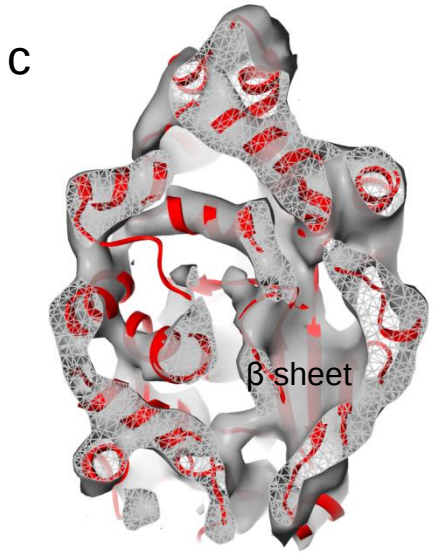
Movie S6, related to Fig. 5. Triplex heterotrimer.

Movie S7, related to Fig. 6. Interactions between triplex heterotrimer and floor regions of hexons.

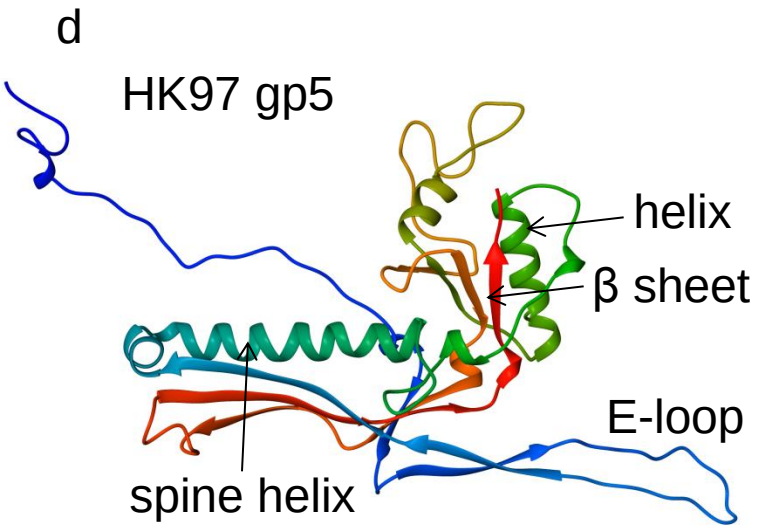
Movie S8, related to Fig. 7. Model of maturation.

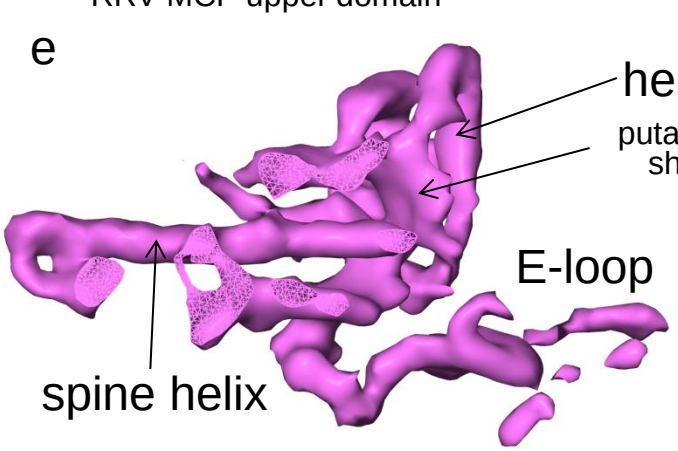




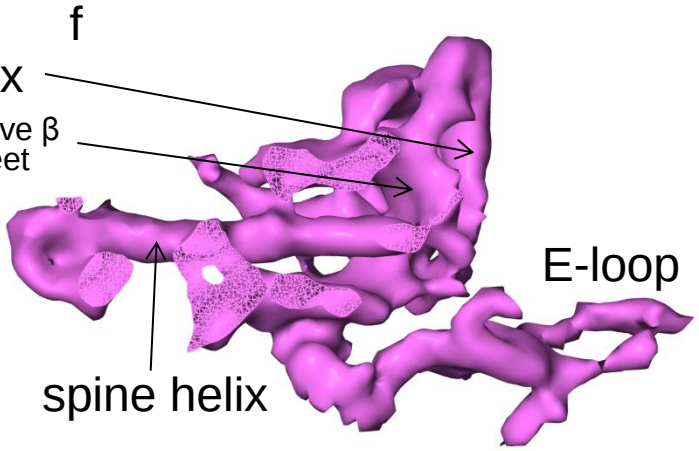


RRV MCP upper domain



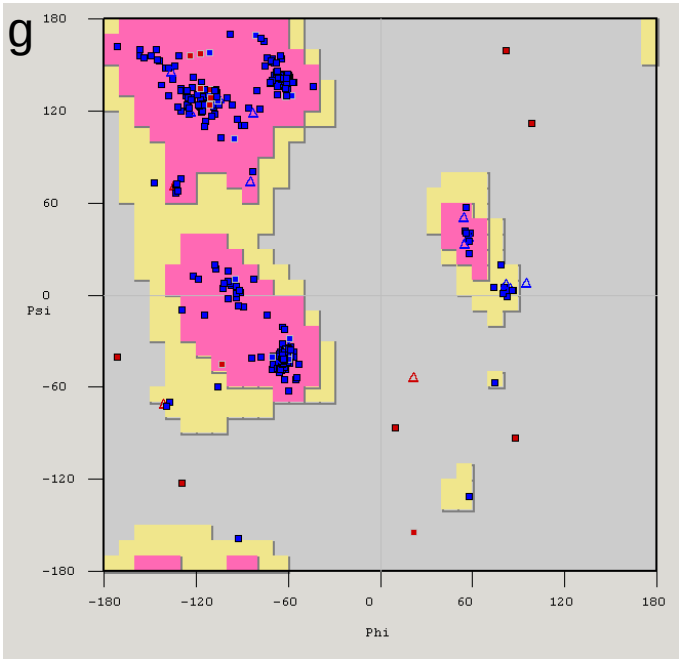


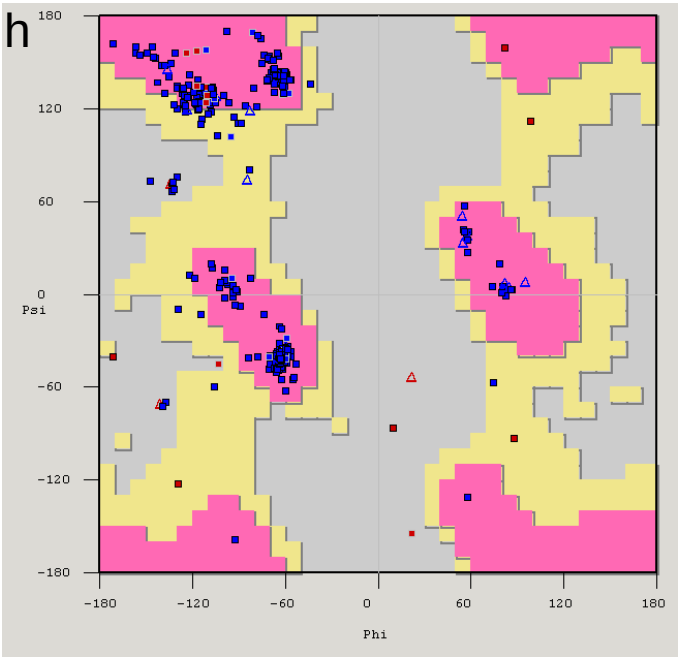
RRV MCP Johnson-fold domain
(high threshold display)



RRV MCP Johnson-fold domain
(low threshold display)

Supplementary Fig. S1 a-f





Preferred regions: 513 (92.93%; Allowed regions: 21 (3.80%); Outliers: 18 (3.26%)

Figure S1, Related to Fig. 1. Assessment of resolution and validation of secondary structures of the cryoEM reconstruction. (**a**) "Gold standard" Fourier shell correlation (FSC) coefficient between two maps, each independently processed and reconstructed of half of the full dataset. The effective resolution reached 7.2 Å based on the 0.143 gold standard FSC criterion. (**b**) A central density slice of the cryoEM map shows the protein capsid shell (bright) and the flat, featureless background away from the capsid shell. (**c-f**) Identification of secondary structure elements in the final 3D map of RRV capsid. (**c**) A cut-away view of the upper domain of RRV MCP subunit superimposed with its pseudo-atomic model derived from the crystal structure of HSV-1 MCPud (Bowman et al., 2003) (red ribbon). Note that the β sheet resolved in the interior of the density map corresponds to that in the pseudo-atomic model. (**d-f**) The ribbon model of bacteriophage HK97 gp5 (**d**) is shown in the same orientation as the shaded surface view of a slab of the Johnson-fold domain of the RRV MCP (**e** and **f**), which was segmented out from the cryoEM reconstruction of the RRV capsid. The RRV Johnson-fold is displayed at two different density thresholds, a high density threshold suitable for visualization of α helices and β sheets (**e**) and a low density threshold suitable for visualization of the E-loop structure (**f**). For clarity, the front portion of the RRV Johnson-fold domain is made invisible with the clip plane tool of the Chimera visualization software in (**e**) and (**f**). (**g-h**) Ramachandran plots of the MCP upper domain for non-Pro/non-Gly residues and Pro/Gly residues. Squares and triangles represent non-Pro/non-Gly and pro/gly residues, respectively. Blue indicates residues that lie within preferred and allowed regions, whereas red indicates residues that fall in outlier regions. Pink, yellow, and gray areas denote preferred, allowed, and outlier Ramachandran values for non-Pro/non-Gly residues and for Pro/Gly residues in (**g**) and (**h**), respectively.

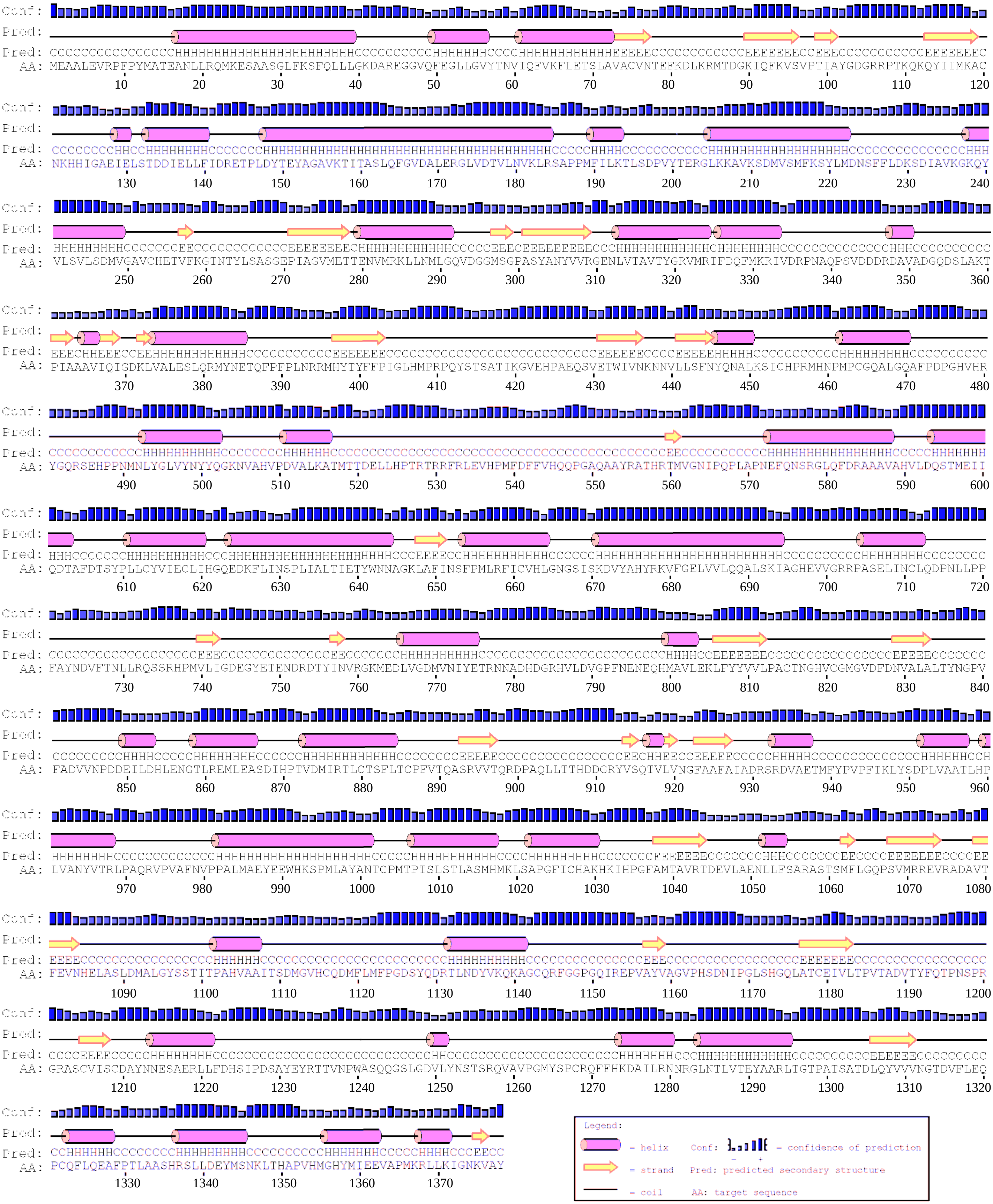


Figure S2, Related to Fig. 3. Secondary structure prediction of MCP. Confidence level (Conf) is displayed by blue tic marks and sequence numbers are indicated by the numbers beneath the sequence. α helices, β strands and loops are shown as pink cylinders, yellow arrows and black lines, respectively.

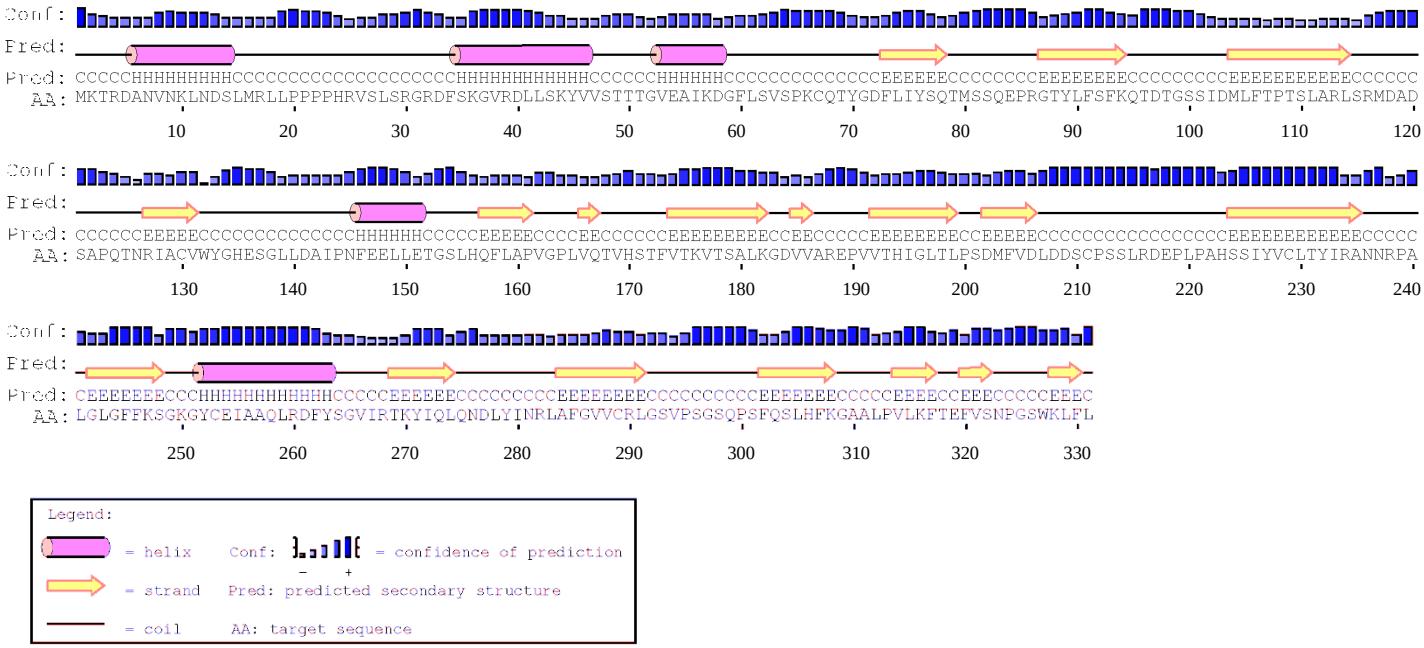


Figure S3, Related to Fig. 5. Predicted secondary structure elements of TRI-1. Confidence level (Conf) is displayed by blue tic marks, and sequence numbers are indicated by the numbers beneath the sequence. α helices, β strands and loops/coils are shown as pink cylinders, yellow arrows and black lines, respectively.

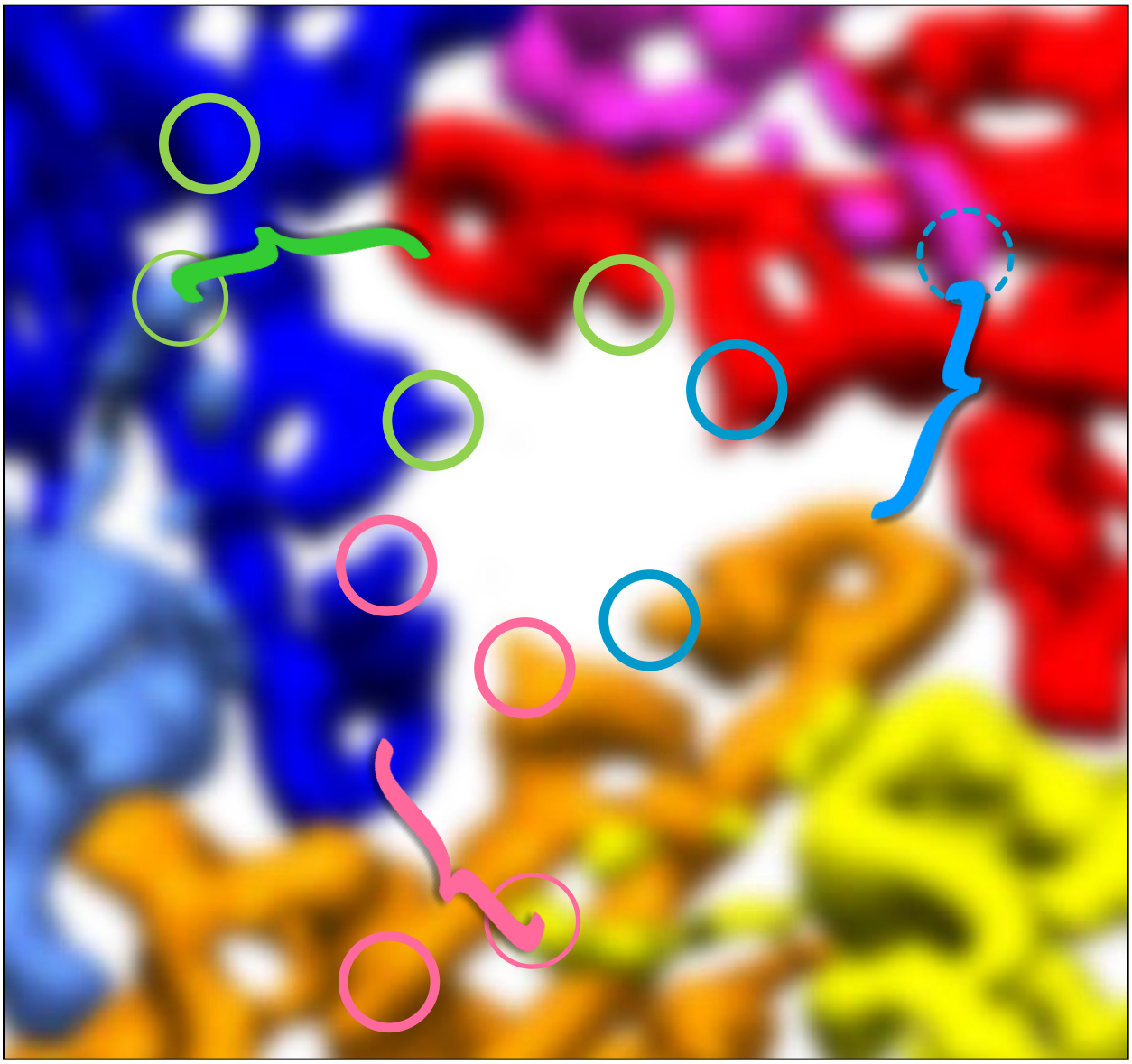


Figure S4, Related to Fig. 2d and Fig. 6g. Non-covalent interactions mediated by the clamp domain of triplex monomers. Each curly brace denotes the interactions of a triplex monomer (pink, cyan or green, following **Figs. 2d, 4** and **6g**) that joins the E loop of the Johnson-fold domain in an outer MCP to the P subdomain of the Johnson-fold domain of an inner MCP, thus outer yellow to inner blue, outer magenta to inner orange, and outer light blue to inner red. These non-covalent interactions stand in place of the covalent link between the analogous E-loop and P-domain of HK97 chainmail.