

## Supplementary Materials for

### Structure of native HIV-1 cores and their interactions with IP6 and CypA

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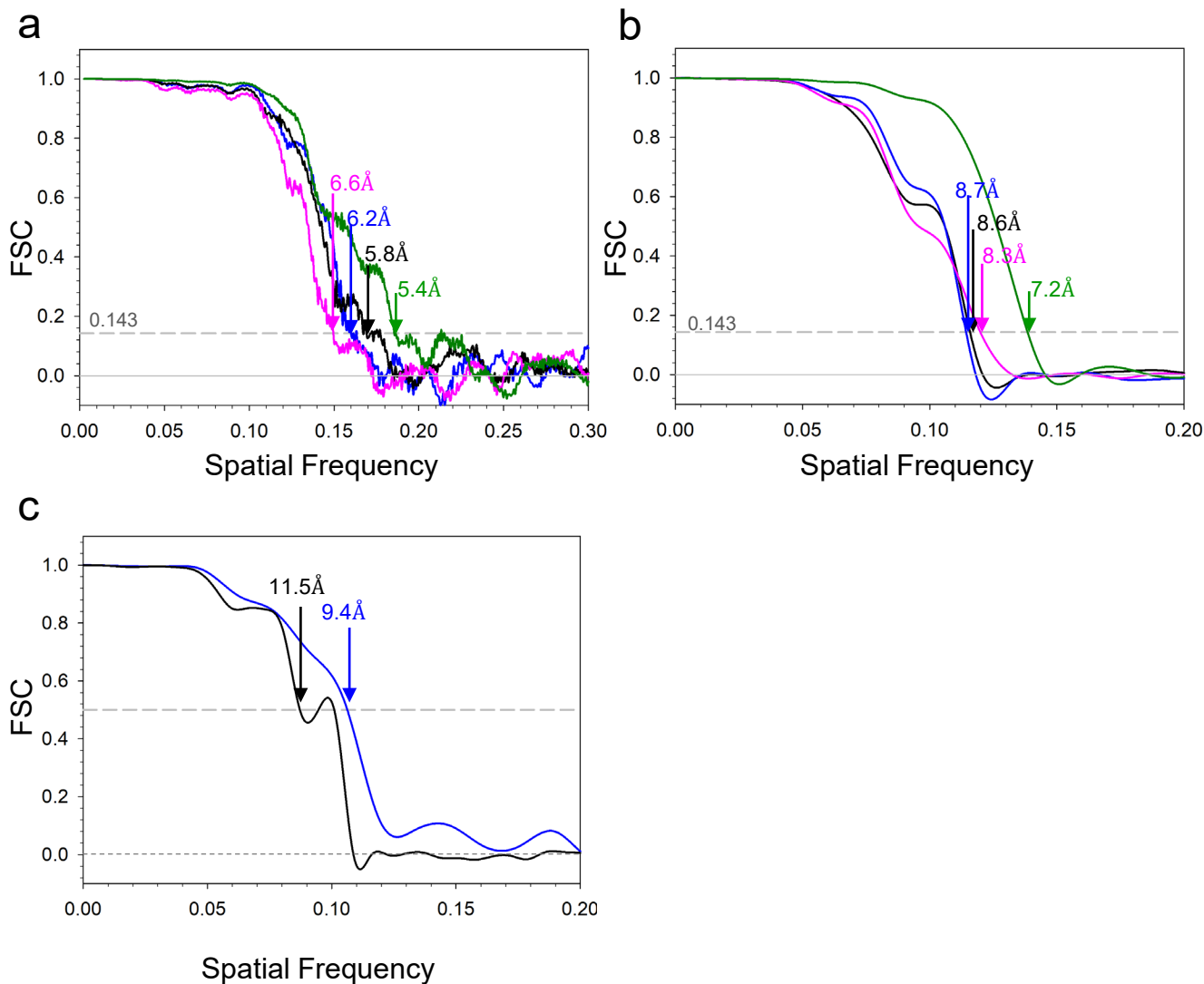
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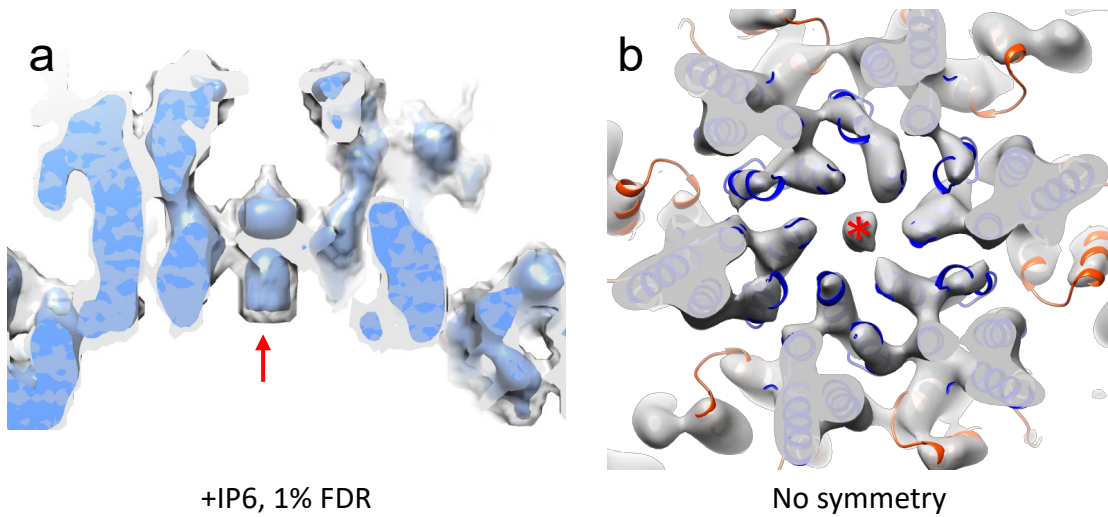
Table S1  
Figs. S1 to S4

**Table S1.** Comparison of subtomogram averaging using PFO-treated virions and those without treatment(Mattei et al. (ref 24)).

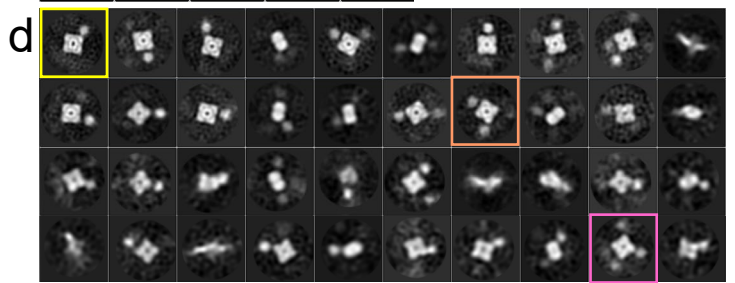
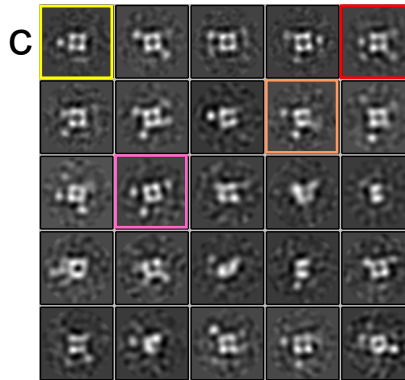
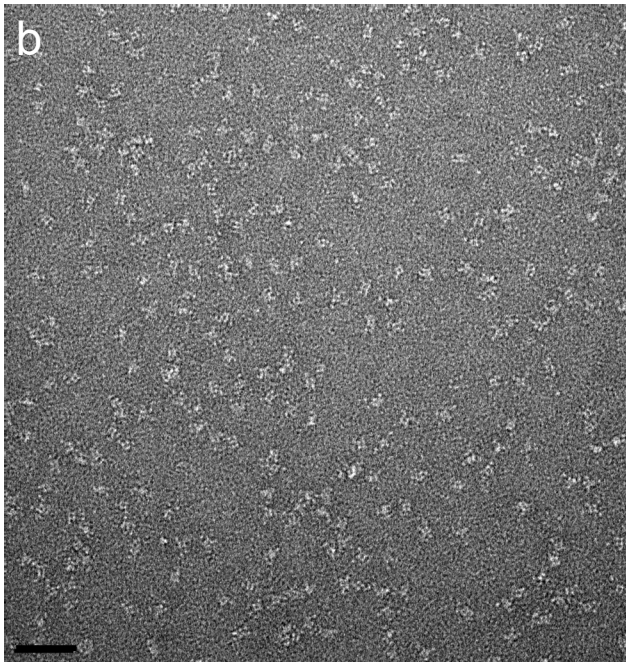
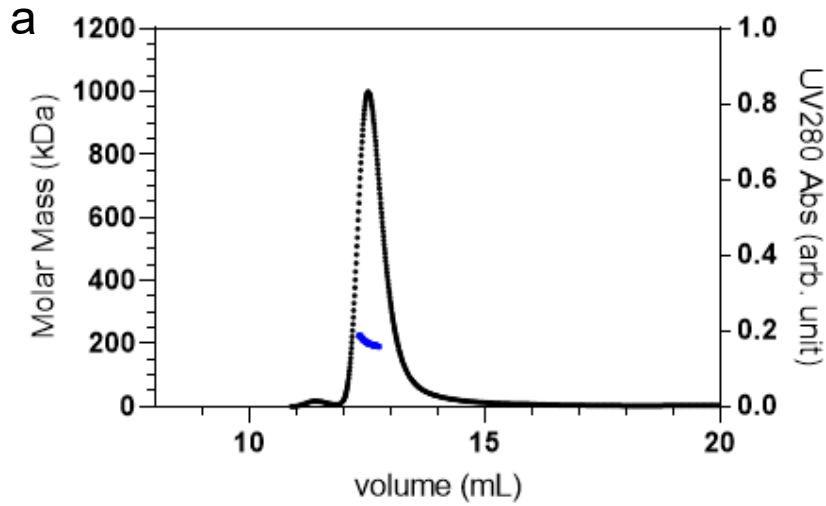
	# of tilt series	# of cores	# of subtomograms	Pixel size (Å)	Defocus (μm)	Resolution (Å)
Cores Mattei et al	103	552	72,836	1.78	-2 to -6.5	6.8
PFO-Cores This study	101	157	32,114	1.04	-2 to -3	5.8
PFO-Cores + IP6 This study	109	481	82,837	1.06	-2.5 to -7	5.4



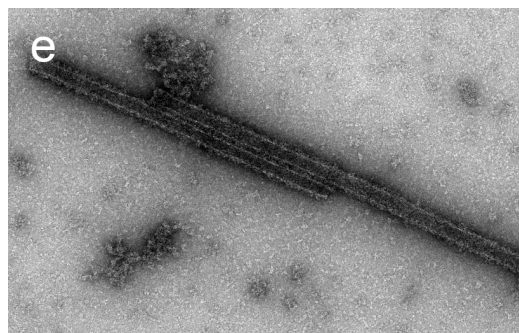
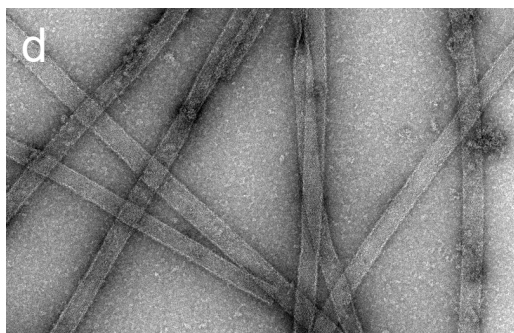
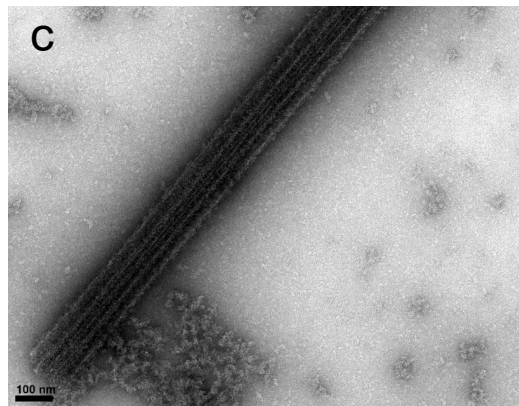
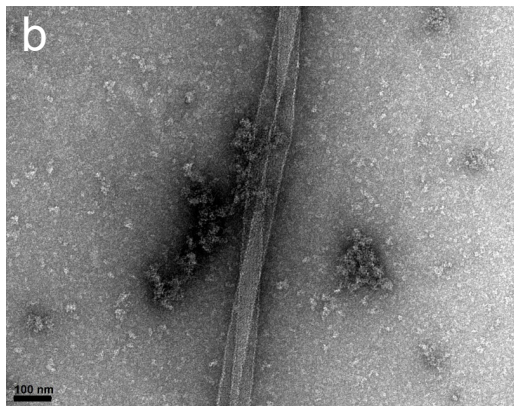
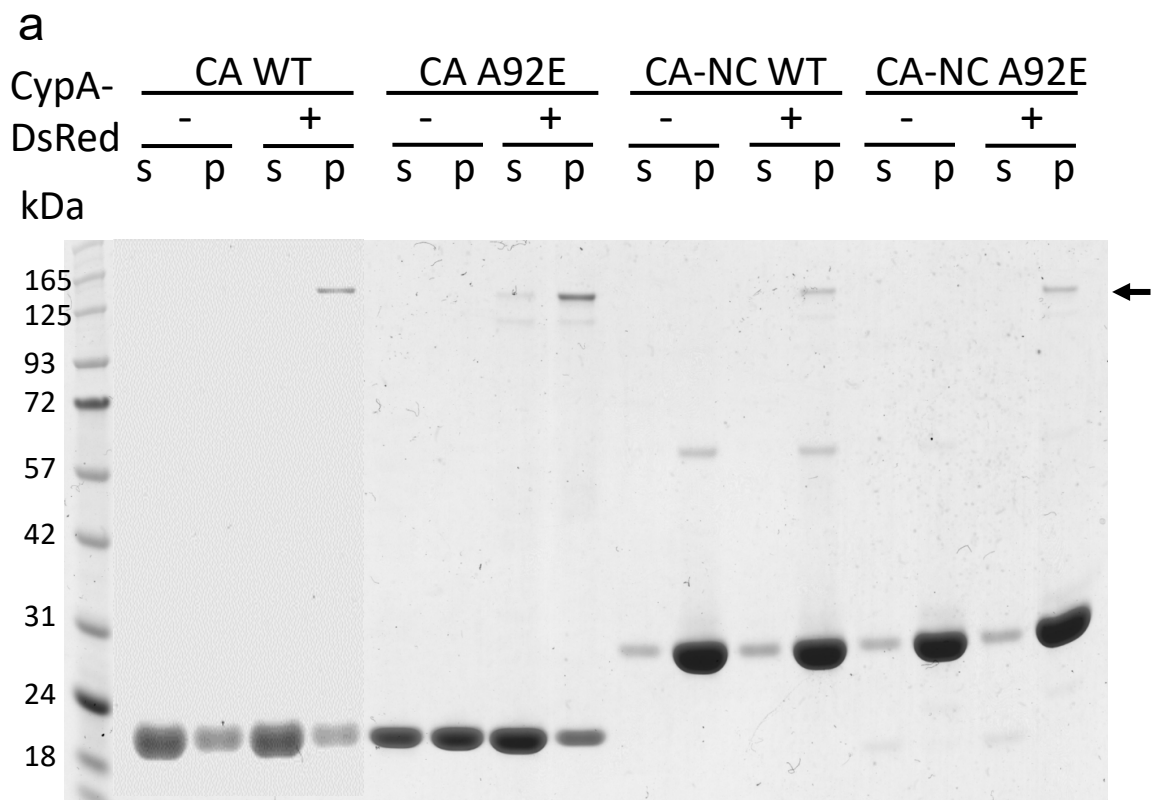
**Figure S1 | Fourier Shell Correlations (FSC) of HIV-1 capsid capsomer maps.** a) FSC plots of CA hexamers in the apo state (black), in the presence of IP6/CypA (blue), IP6/CypA-DsRed (pink). b) FSC plots of CA pentamers in the apo state (black), in the presence of IP6/CypA (blue) and IP6/CypA-DsRed (pink). c) FSC plots of CypA-DsRed in complex with CA hexamers from subtomogram classes of one CypA-DsRed (black) and two CypA-DsRed (blue) bound to CA hexamers. A soft mask enclosing the central hexamer was used during FSCs calculation.



**Figure S2 | Analysis of the central density in IP6/CA hexamer and IP6/CypA-DsRed/CA hexamer complexes.** a) Side view of confidence map at 1% false discovery rate (transparent gray) overlaid with IP6/CA hexamer map (blue). b) Subtomogram averaging of IP6/CypA-DsRed/CA hexamer without applying six-fold symmetry throughout the processing. The red asterisk and arrow indicate the central density.



**Figure S3 | Purification and characterization of CypA-DsRed.** a) Multi-angle light scattering of CypA-DsRed. The protein at 3.0 mg/mL was injected into an analytical Superdex 200 gel filtration column at a flow rate of 0.5 ml/min. Molecular mass of CypA-DsRed (UV absorbance at 280 nm in black solid circle) at the peak elution is indicated with blue solid circle. The estimated average molecular mass of CypA-DsRed across the peak is 201 kDa. b) A negative stain image of purified CypA-DsRed. c-d) 2D classes of CypA-DsRed particles from negative stained images (c) and cryoEM images recorded with a phase plate (d). Yellow, orange, pink and red boxed classes display representative one, two, three and four CypA densities respectively in CypA-DsRed tetramer particles. Scale bar, 50 nm



**Figure S4 | Binding of CypA-DsRed to capsid assemblies.** a) SDS-PAGE analysis of CypA-DsRed binding to WT and A92E CA tubes and CA-NC tubes following pelleting assay, stained with Coomassie Blue. CypA-DsRed runs as a tetramer, marked by an arrow. b-e) Negative stain images of reaction mix from (a) for WT CA assemblies in the absence (b) and presence (c) of CypA-DsRed. d-e) Negative stain images of reaction mix from (a) for A92E CA assemblies in the absence (d) and presence (e) of CypA-DsRed. Scale bars, 100 nm