Supplementary Information

Amyloid fibril structure of islet amyloid polypeptide by cryo-electron microscopy reveals similarities with amyloid beta

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Supplementary Tables

Supplementary Table 1: Data collection for IAPP

Data Collection	
Microscope	Tecnai Arctica
Camera	Falcon 3 (counting mode)
Acceleration voltage (kV)	200
Magnification	110,000
Defocus range (μm)	-1.0 to -2.2
Dose rate (e$^-$/Å^2/s)	0.9
Exposure time (s)	46
Number of movie frames	1800
Pixel size (Å)	0.935

Supplementary Table 2: Reconstruction, modeling and refinement of polymorph 1 (PM1).

	Polymorph 1
Reconstruction	
Box size (pixel)	220
No. fibrils in final reconstruction	1161
No. segments in final reconstruction	37,120
Resolution (0.143 criterion) (Å)	4.2
Helical rise (Å)	2.351
Helical twist (º)	178.23
Helical pitch (Å)	479.5
Symmetry	C1
Model Composition	
Non-hydrogen atoms	2975
Number of chains	16
Model Refinement	
Map CC (around atoms)	0.9018
RMSD bonds (Å)	0.0039
RMSD angles (º)	0.60
All-atom clash score	15.2
Ramachandran outliers/favored (%)	0 / 95.7
Rotamer outliers (%)	0
C-beta deviations	0
Molprobity score	1.99

Supplementary Table 3: Reconstruction of polymorphs 2 (PM2) and 3 (PM3).

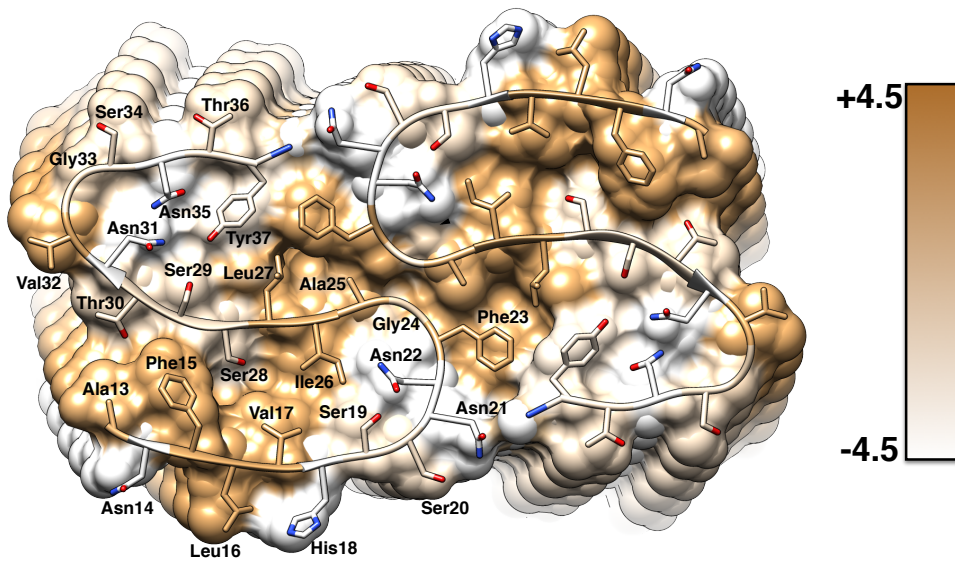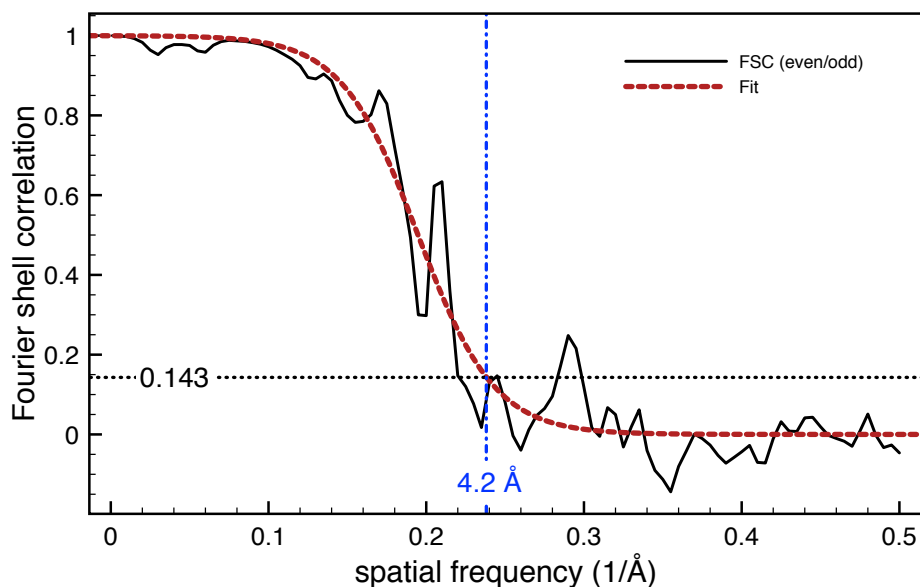|	Polymorph 2	Polymorph 3
Reconstruction		
Box size (pixel)	200	220
No fibrils in final reconstruction	1480	99
No segments in final reconstruction	24011	4591
Resolution (0.143 criterion) (Å)	4.2	8.1
Helical rise (Å)	2.352	2.323
Helical twist (°)	179.10	179.47
Helical pitch (Å)	940	1590
Symmetry	C1 (approx. 2_1 screw)	C1 (approx. 2_1 screw)

Supplementary Figures
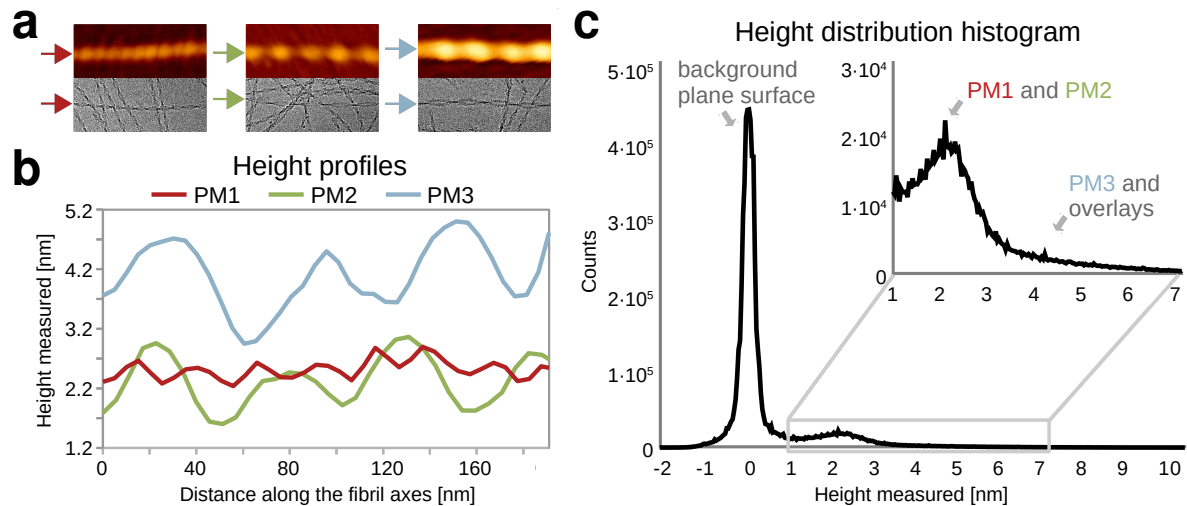
Supplementary Figure 1: Comparison of described IAPP polymorphs. a Single fibril cut-outs of polymorphs PM1, PM2 and PM3 from AFM images (top row) and cryo-EM micrographs (bottom row); single box size is 100x250 nm. **b** Height profiles of individual fibrils extracted from AFM images. **c** Height distribution histogram, showing the highest number of counts for the plane background surface around 0 nm and a distinct peak around 2.2 nm. The peak around 2.2 nm includes both PM1 and PM2 which are non-distinguishable in sense of height distribution. Moreover, a pronounced shoulder on the right corresponds to the presence of lower amounts of PM3 as well as the overlaps of single PM1/PM2 fibrils. For the height distribution analysis, histograms from six height images of 5x5 µm size and a resolution of 1024x1024 pixels were obtained, binned and presented in one graph. An example of the image used can be seen in Supplementary Figure 2.
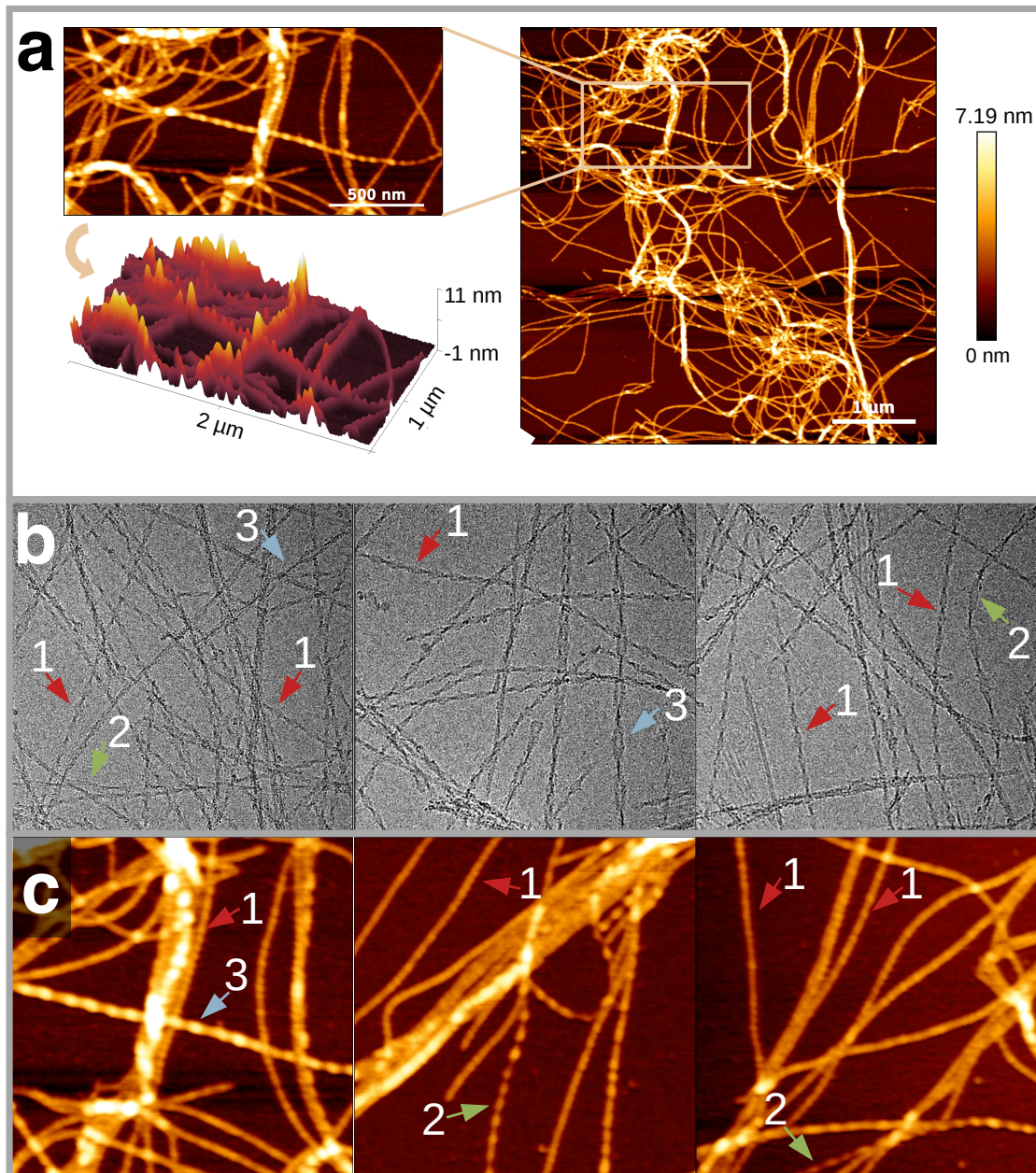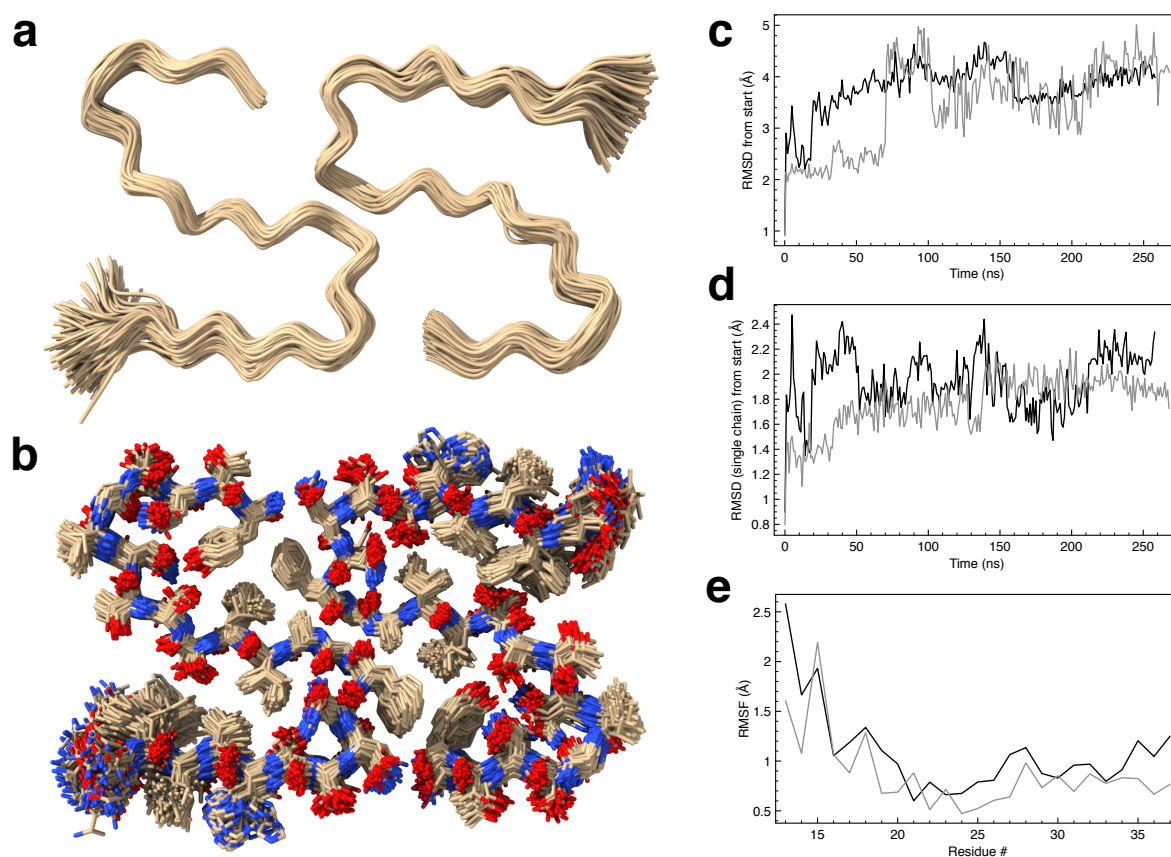
Supplementary Figure 2: Overview of IAPP polymorphs. **a** Typical height profile AFM image used for polymorph distribution analysis. **b** Cryo-EM micrographs showing 370x370 nm areas. **c** AFM overview images showing 1x1 µm areas. Arrows indicate the presence of PM1 (red), PM2 (green) and PM3 (blue).

Supplementary Figure 3: Hydrophobicity plot of the fibril displayed as top view.
Hydrophobicity levels of the IAPP polymorph 1 (PM1) fibril are colored according to
Kyte-Doolittle[1] in the hydrophobicity score range -4.5 (white) to 4.5 (gold). One
hydrophobic cluster spans the entire diagonal of the fibril cross-section. This
hydrophobic streak is surrounded by highly ordered polar clusters.

Supplementary Figure 4: FSC Analysis of polymorph 1 (PM1). FSC curves from the even/odd test (solid black) from the gold-standard refinement yields a resolution of 4.2 Å (using the 0.143 criterion). The even/odd FSC curve is fitted (red) with the model function $1/(1+\exp((x-A)/B))$ (with $A=0.1947$ and $B=0.026$) to obtain a more robust resolution estimate.

Supplementary Figure 5: Results of molecular dynamics simulations of IAPP polymorph 1 (PM1). Superimposed snapshots from a 250 ns simulation displaying only the backbone (**a**) or all atoms (except for solvent and hydrogen) (**b**). **c** Showing the RMSD from the deposited structure of PM1 (PDB ID 6Y1A) for two 250 ns simulations (black and grey lines, respectively). **d** Showing the RMSD of a single chain from the deposited structure during the two 250 ns simulations. **e** Showing the atomic root mean square fluctuations (RMSF) for each residue calculated over each 250 ns simulation.

#	Cwork	Cfree	Sequence (residues 1–21 boxed)
1	0.845	0.180	KCNTATCATQRLAN·FLVHSSNNFGAILSSTNVGSN·TY
2	0.807	0.151	KC·NTATCATQRLANFLVHSSNNF·GAILSSTNVGSNTY
3	0.818	0.133	KCNTATCATQRLA·NFLVHSSNNFGAILSSTNVGS·NTY
4	0.828	0.128	KCNTATCATQR·LANFLVHSSNNFGAILSSTNV·GSNTY
5	0.815	0.125	KCNTATCATQRLANF·LVHSSNNFGAILSSTNVGSNTY·Y
6	0.798	0.124	KCNTATC·ATQRLANFLVHSSNNFGAILS·STNVGSNTY
7	0.768	0.121	YTNS·GVNTSSLIAGFNNSSHVLFNA·LRQTACTATNCK
8	0.755	0.118	YTNSGVN·TSSLIAGFNNSSHVLFNALRQ·TACTATNCK
9	0.789	0.115	YTNSGVNT·SSLIAGFNNSSHVLFNALRQT·ACTATNCK
10	0.832	0.111	KCNTATCATQ·RLANFLVHSSNNFGAILSSTN·VGSNTY
11	0.798	0.110	KCNTATCATQRLANFL·VHSSNNFGAILSSTNVGSNTY
12	0.811	0.109	YTNSGVNTSSL·IAGFNNSSHVLFNALRQTACT·ATNCK
13	0.796	0.101	KCNTATCA·TQRLANFLVHSSNNFGAILSS·TNVGSNTY
14	0.751	0.095	Y·TNSGVNTSSLIAGFNNSSHVL·FNALRQTACTATNCK
15	0.771	0.095	YTNSGV·NTSSLIAGFNNSSHVLFNALR·QTACTATNCK
16	0.780	0.094	YTN·SGVNTSSLIAGFNNSSHVLFN·ALRQTACTATNCK
17	0.813	0.094	KCN·TATCATQRLANFLVHSSNNFG·AILSSTNVGSNTY
18	0.817	0.090	KCNTA·TCATQRLANFLVHSSNNFGAI·LSSTNVGSNTY
19	0.763	0.085	YT·NSGVNTSSLIAGFNNSSHVLF·NALRQTACTATNCK
20	0.807	0.082	KCNTATCAT·QRLANFLVHSSNNFGAILSST·NVGSNTY
21	0.799	0.079	KCNT·ATCATQRLANFLVHSSNNFGA·ILSSTNVGSNTY
22	0.781	0.072	YTNSGVNTSSLIA·GFNNSSHVLFNALRQTACTAT·NCK
23	0.779	0.071	YTNSGVNTSSLIAGFN·NSSHVLFNALRQTACTATNCK
24	0.766	0.071	YTNSGVNTSSLIAGFNNSSHVL·FNALRQTACTATNCK
25	0.795	0.069	K·CNTATCATQRLANFLVHSSNN·FGAILSSTNVGSNTY
26	0.758	0.063	YTNSGVNTS·SLIAGFNNSSHVLFNALRQTA·CTATNCK
27	0.825	0.058	KCNTAT·CATQRLANFLVHSSNNFGAIL·SSTNVGSNTY
28	0.792	0.054	KCNTATCATQRL·ANFLVHSSNNFGAILSSTNVG·SNTY
29	0.800	0.054	KCNTATCATQRLANFLVHSSN·NFGAILSSTNVGSNTY
30	0.763	0.050	YTNSG·VNTSSLIAGFNNSSHVLFNAL·RQTACTATNCK
31	0.743	0.045	YTNSGVNTSSLIAG·FNNSSHVLFNALRQTACTATN·CK
32	0.754	0.042	YTNSGVNTSS·LIAGFNNSSHVLFNALRQTAC·TATNCK
33	0.758	0.033	YTNSGVNTSSLIAGF·NNSSHVLFNALRQTACTATNC·K
34	0.757	0.025	YTNSGVNTSSLI·AGFNNSSHVLFNALRQTACTA·TNCK
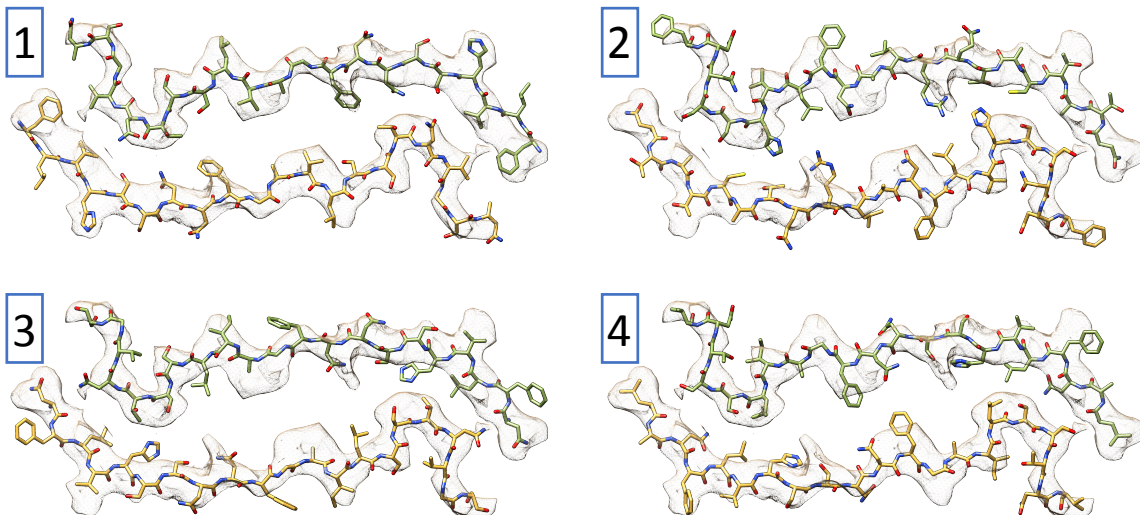
Supplementary Figure 6: DireX² analysis of polymorph 2 (PM2). Ranking of the potential IAPP sequence snippets that have been modeled into the reconstructed density according to their C_{free} values. Each snippet contains 21 amino acid residues which were modeled into the density map in both possible Cα-chain directions. Atomic models of the four most favorable sequence snippets are shown at the bottom. Note that some models, for example model 2, can be excluded since they are incompatible with the disulfide bond between residues Cys[2] and Cys[7].
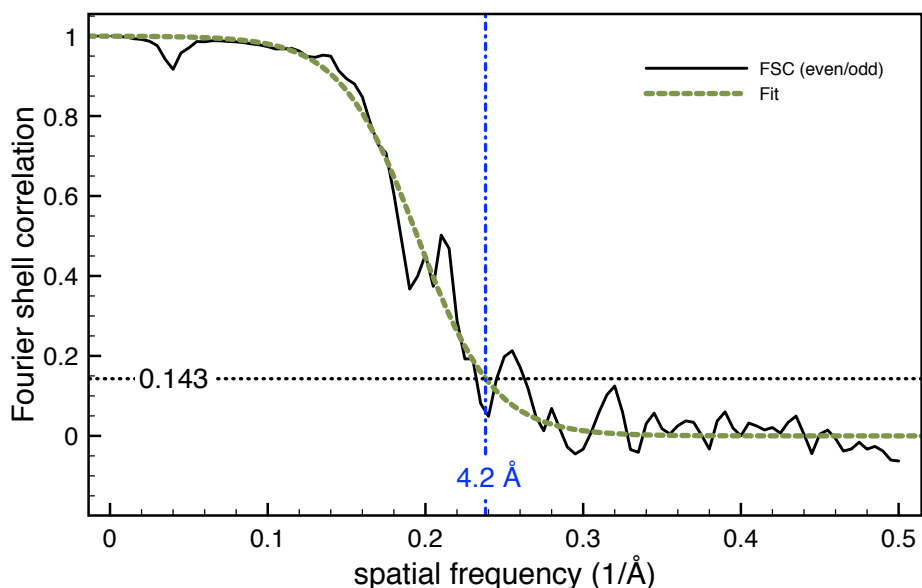
Supplementary Figure: 7 FSC analysis of polymorph 2 (PM2). FSC curves from the even/odd test (solid black) from the gold-standard refinement yields a resolution of 4.2 Å (using the 0.143 criterion). The even/odd FSC curve is fitted (green) with the model function $1/(\,1+\exp((x-A)/B))\,)$ (with A=0.194789 and B=0.02427) to obtain a more robust resolution estimate.
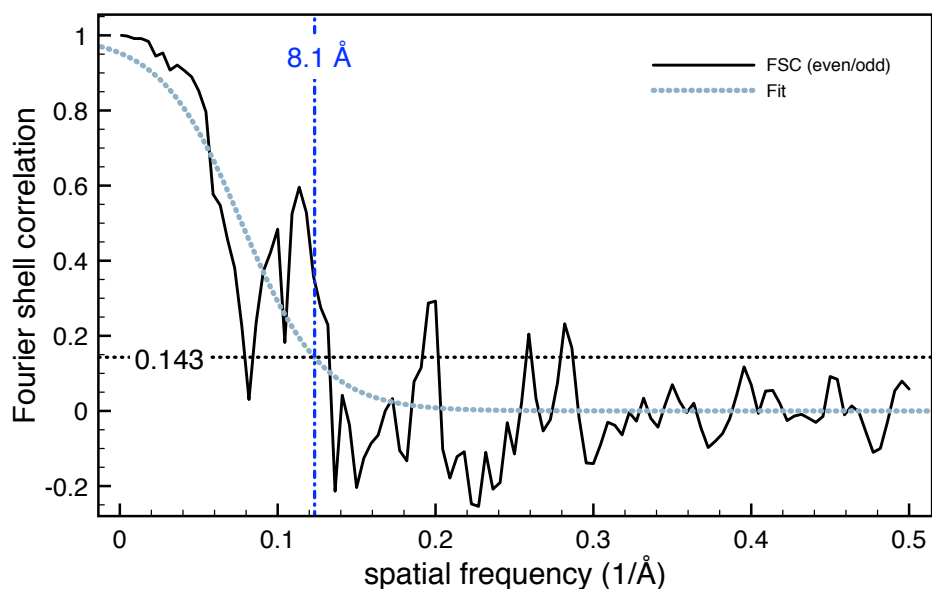
Supplementary Figure 8: FSC analysis of Polymorph 3 (PM3). FSC curves from the even/odd test (solid black) from the gold-standard refinement yields a resolution of 8.1 Å (using the 0.143 criterion). The even/odd FSC curve is fitted (light blue) with the model function $1/(\,1+\exp((x-A)/B)\,)$ (with A=0.0772 and B=0.0256) to obtain a more robust resolution estimate.