wwPDB EM Validation Summary Report

Nov 29, 2022 – 03:55 PM EST

PDB ID	:	8DCP
EMDB ID	:	EMD-27327
Title	:	PI 3-kinase alpha with nanobody 3-126
Authors	:	Hart, J.R.; Liu, X.; Pan, C.; Liang, A.; Ueno, L.; Xu, Y.; Quezada, A.; Zou, X.; Yang, S.; Zhou, Q.; Schoonooghe, S.; Hassanzadeh-Ghassabeh, G.; Xia, T.; Shui, W.; Yang, D.; Vogt, P.K.; Wang, M.-W.
Deposited on	:	2022-06-17
Resolution	:	2.41 Å(reported)
Based on initial model	:	7MYN

This is a wwPDB EM Validation Summary Report for a publicly released PDB entry.

We welcome your comments at *validation@mail.wwpdb.org*
A user guide is available at
https://www.wwpdb.org/validation/2017/EMValidationReportHelp
with specific help available everywhere you see the ⓘ symbol.

The types of validation reports are described at
http://www.wwpdb.org/validation/2017/FAQs#types.

The following versions of software and data (see references ⓘ) were used in the production of this report:

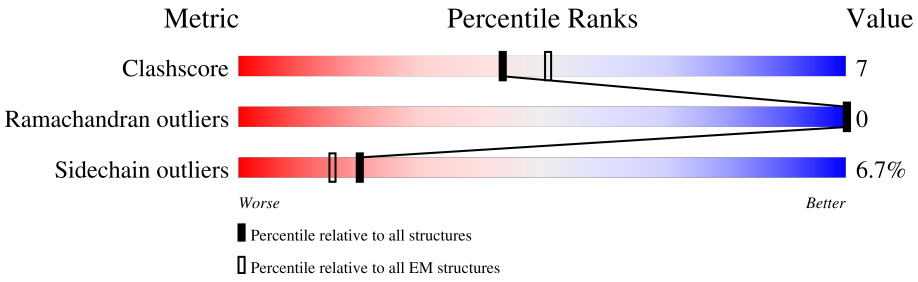
EMDB validation analysis	:	0.0.1.dev43
MolProbity	:	4.02b-467
Percentile statistics	:	20191225.v01 (using entries in the PDB archive December 25th 2019)
MapQ	:	1.9.9
Ideal geometry (proteins)	:	Engh & Huber (2001)
Ideal geometry (DNA, RNA)	:	Parkinson et al. (1996)
Validation Pipeline (wwPDB-VP)	:	2.31.2

1 Overall quality at a glance (i)

The following experimental techniques were used to determine the structure:
ELECTRON MICROSCOPY

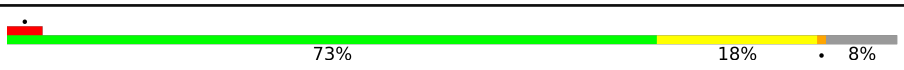
The reported resolution of this entry is 2.41 Å.

Percentile scores (ranging between 0-100) for global validation metrics of the entry are shown in the following graphic. The table shows the number of entries on which the scores are based.



Metric	Whole archive (#Entries)	EM structures (#Entries)
Clashscore	158937	4297
Ramachandran outliers	154571	4023
Sidechain outliers	154315	3826

The table below summarises the geometric issues observed across the polymeric chains and their fit to the map. The red, orange, yellow and green segments of the bar indicate the fraction of residues that contain outliers for >=3, 2, 1 and 0 types of geometric quality criteria respectively. A grey segment represents the fraction of residues that are not modelled. The numeric value for each fraction is indicated below the corresponding segment, with a dot representing fractions <=5% The upper red bar (where present) indicates the fraction of residues that have poor fit to the EM map (all-atom inclusion < 40%). The numeric value is given above the bar.

Mol	Chain	Length	Quality of chain
1	A	1096	
2	B	723	

2 Entry composition (i)

There are 3 unique types of molecules in this entry. The entry contains 10595 atoms, of which 0 are hydrogens and 0 are deuteriums.

In the tables below, the AltConf column contains the number of residues with at least one atom in alternate conformation and the Trace column contains the number of residues modelled with at most 2 atoms.

- Molecule 1 is a protein called Phosphatidylinositol 4,5-bisphosphate 3-kinase catalytic subunit alpha isoform.

Mol	Chain	Residues	Atoms					AltConf	Trace
1	A	1005	Total	C	N	O	S	1	0
			8233	5260	1415	1491	67		

There are 28 discrepancies between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	Comment	Reference
A	-27	MET	-	expression tag	UNP P42336
A	-26	SER	-	expression tag	UNP P42336
A	-25	TYR	-	expression tag	UNP P42336
A	-24	TYR	-	expression tag	UNP P42336
A	-23	HIS	-	expression tag	UNP P42336
A	-22	HIS	-	expression tag	UNP P42336
A	-21	HIS	-	expression tag	UNP P42336
A	-20	HIS	-	expression tag	UNP P42336
A	-19	HIS	-	expression tag	UNP P42336
A	-18	HIS	-	expression tag	UNP P42336
A	-17	ASP	-	expression tag	UNP P42336
A	-16	TYR	-	expression tag	UNP P42336
A	-15	ASP	-	expression tag	UNP P42336
A	-14	ILE	-	expression tag	UNP P42336
A	-13	PRO	-	expression tag	UNP P42336
A	-12	THR	-	expression tag	UNP P42336
A	-11	THR	-	expression tag	UNP P42336
A	-10	GLU	-	expression tag	UNP P42336
A	-9	ASN	-	expression tag	UNP P42336
A	-8	LEU	-	expression tag	UNP P42336
A	-7	TYR	-	expression tag	UNP P42336
A	-6	PHE	-	expression tag	UNP P42336
A	-5	GLN	-	expression tag	UNP P42336
A	-4	GLY	-	expression tag	UNP P42336
A	-3	ALA	-	expression tag	UNP P42336
A	-2	MET	-	expression tag	UNP P42336
A	-1	GLY	-	expression tag	UNP P42336

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Chain	Residue	Modelled	Actual	Comment	Reference
A	0	SER	-	expression tag	UNP P42336

- Molecule 2 is a protein called Phosphatidylinositol 3-kinase regulatory subunit alpha.

Mol	Chain	Residues	Atoms					AltConf	Trace
2	B	276	Total	C	N	O	S	0	0
			2341	1464	420	449	8		

There is a discrepancy between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	Comment	Reference
B	2	MET	-	initiating methionine	UNP P27986

- Molecule 3 is water.

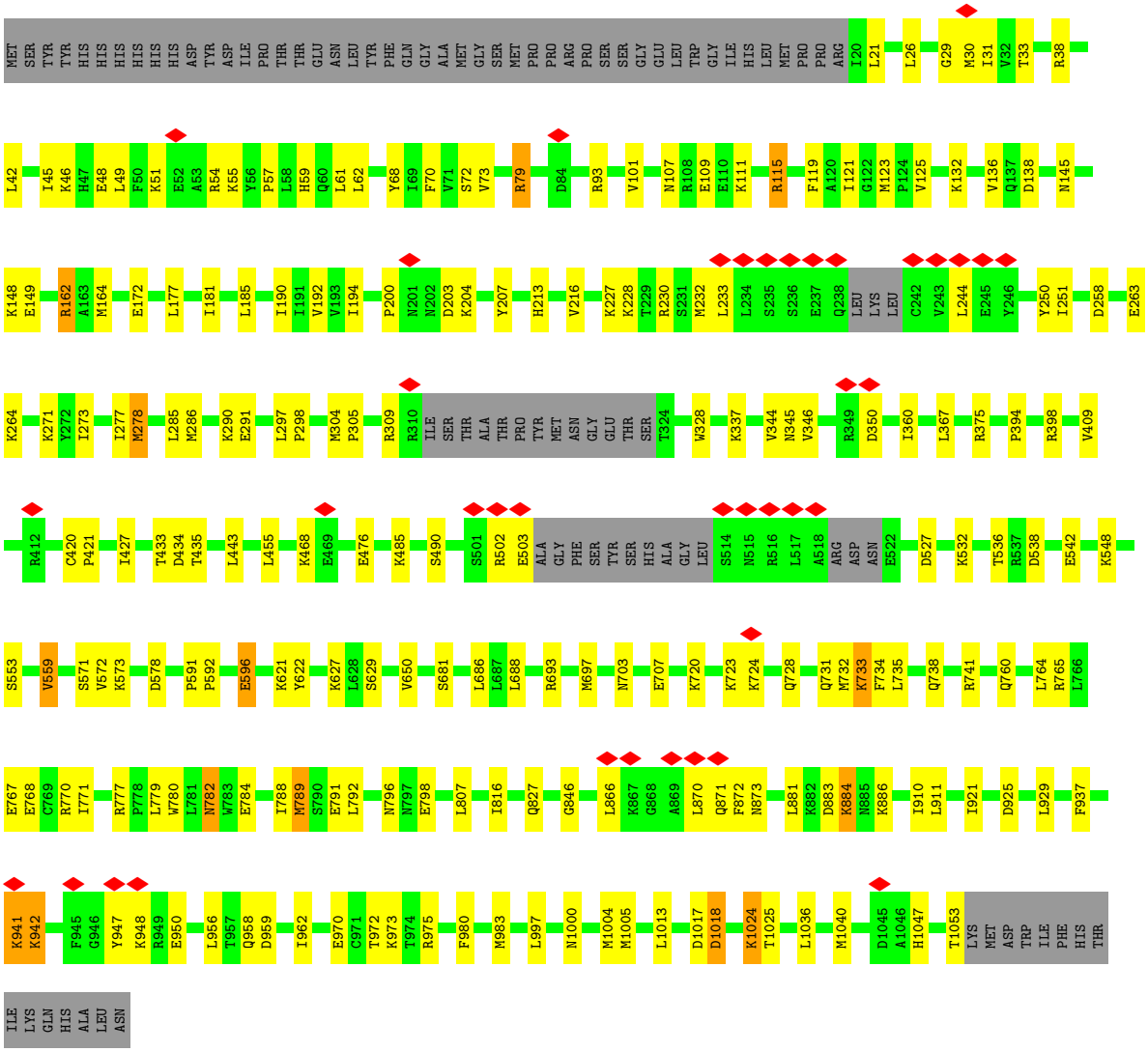
Mol	Chain	Residues	Atoms		AltConf
3	A	20	Total	O	0
			20	20	
3	B	1	Total	O	0
			1	1	

3 Residue-property plots (i)

These plots are drawn for all protein, RNA, DNA and oligosaccharide chains in the entry. The first graphic for a chain summarises the proportions of the various outlier classes displayed in the second graphic. The second graphic shows the sequence view annotated by issues in geometry and atom inclusion in map density. Residues are color-coded according to the number of geometric quality criteria for which they contain at least one outlier: green = 0, yellow = 1, orange = 2 and red = 3 or more. A red diamond above a residue indicates a poor fit to the EM map for this residue (all-atom inclusion < 40%). Stretches of 2 or more consecutive residues without any outlier are shown as a green connector. Residues present in the sample, but not in the model, are shown in grey.

• Molecule 1: Phosphatidylinositol 4,5-bisphosphate 3-kinase catalytic subunit alpha isoform

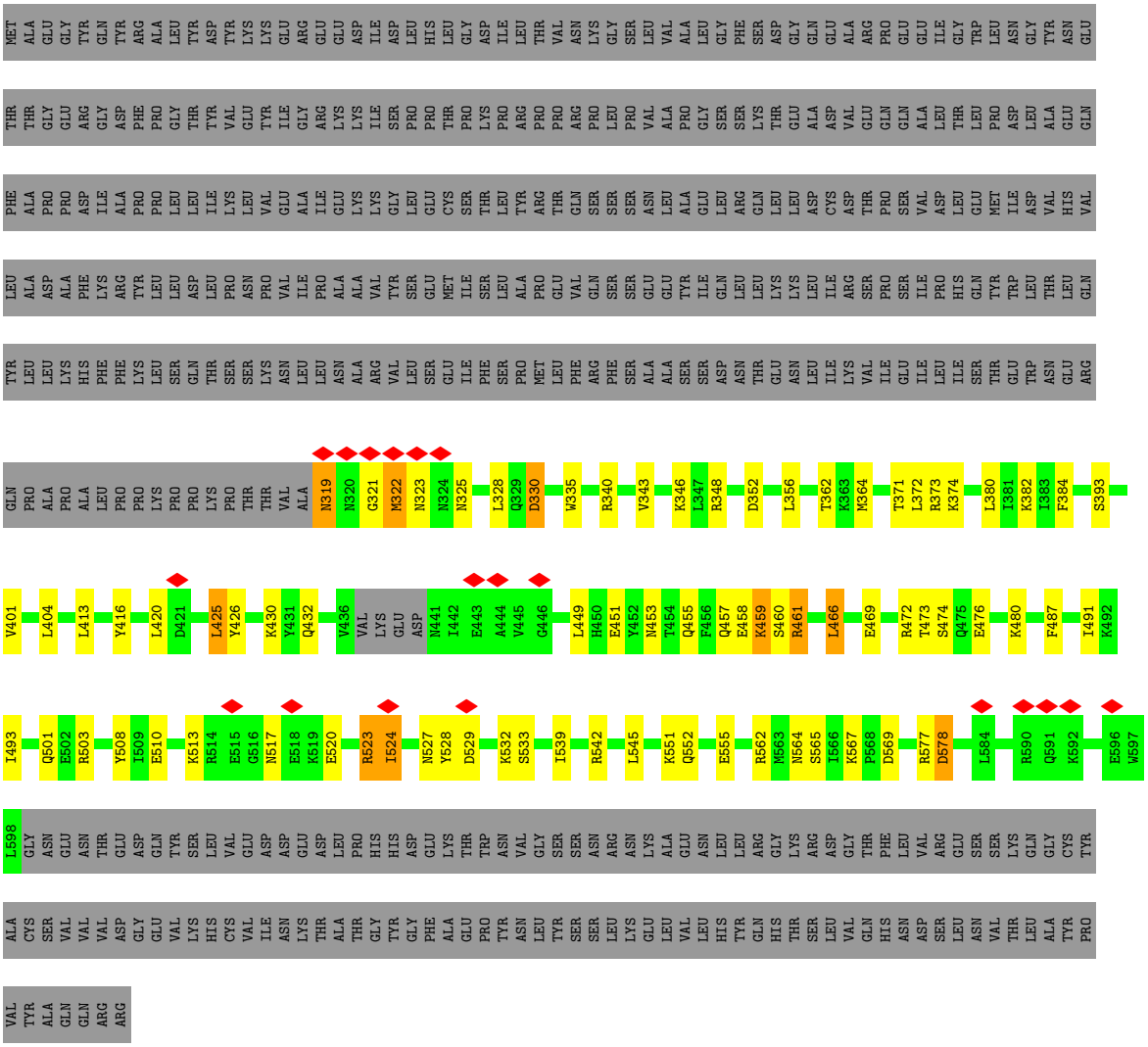
Chain A:



• Molecule 2: Phosphatidylinositol 3-kinase regulatory subunit alpha

Chain B:


27%
10%
62%


MET ALA GLU GLY TYR GLN TYR ARG ALA LEU TYR ASP TYR LYS LYS GLU ARG GLU GLU ASP ILE ASP LEU HIS LEU GLY ASP ILE LEU THR VAL ASN LYS GLY SER LEU VAL ALA ALA LEU GLY PHE SER ASP GLY GLN GLU ALA ARG PRO GLU GLU GLU ILE GLY TRP LEU ASN GLY TYR ASN GLU
THR THR GLY GLU ARG GLY ASP PHE PRO GLY THR TYR VAL GLU TYR ILE GLY ARG LYS LYS ILE SER PRO PRO THR PRO LYS PRO ARG PRO PRO ARG PRO LEU PRO VAL ALA PRO GLY SER SER LYS THR GLU ALA ASP VAL GLU GLN GLN ALA LEU THR LEU PRO ASP LEU ALA GLU GLN
PHE ALA PRO PRO ASP ILE ALA PRO PRO LEU LEU ILE LYS LEU VAL GLU ALA ILE GLU LYS LYS GLY LEU GLU CYS SER THR LEU TYR ARG THR GLN SER SER SER ASN LEU ALA GLU LEU ARG GLN LEU LEU ASP CYS ASP THR PRO SER VAL ASP LEU GLU MET ILE ASP VAL HIS VAL
LEU ALA ASP ALA PHE LYS ARG TYR LEU LEU ASP LEU PRO ASN PRO VAL ILE PRO ALA ALA VAL TYR SER GLU MET ILE SER LEU ALA PRO GLU VAL GLN SER SER GLU GLU TYR ILE GLN LEU LEU LEU LYS LYS LEU ILE ARG SER PRO SER ILE PRO HIS GLN TYR TRP LEU THR LEU GLN
TYR LEU LEU LYS HIS PHE PHE LYS LEU SER GLN THR SER SER LYS ASN LEU LEU ASN ALA ARG VAL LEU SER GLU ILE PHE SER PRO MET LEU PHE ARG PHE SER ALA ALA SER SER ASP ASN THR GLU ASN LEU ILE LYS VAL ILE GLU ILE LEU LEU ILE SER THR GLU TRP ASN GLU ARG
GLN PRO ALA PRO ALA LEU PRO PRO LYS PRO PRO LYS PRO THR THR VAL ALA N319 M320 G321 M322 N323 N324 N325 L328 Q329 D330 W335 R340 V343 K346 L347 R348 D352 L356 T362 K363 M364 T371 L372 R373 K374 L380 I381 K382 I383 F384 S393
V401 L404 L413 Y416 L420 D421 L425 Y426 K430 Y431 Q432 V436 VAL LYS GLU ASP N441 I442 E443 A444 V445 G446 L449 H450 E451 V452 N453 T454 Q455 F456 Q457 E458 K459 S460 R461 L466 E469 R472 T473 S474 Q475 E476 K480 F487 I491 K492
I493 Q501 E502 R503 Y508 I509 E510 K513 R514 E515 G516 N517 E518 K519 E520 R523 I524 N527 Y528 D529 K532 S533 I539 R542 L545 K551 Q552 E555 R562 M563 N564 S565 I566 K567 P568 D569 R577 D578 L584 R590 Q591 K592 E596 W597
L598 GLY ASN GLU ASN THR GLU ASP GLN TYR SER LEU VAL GLU ASP ASP GLU ASP LEU PRO HIS HIS ASP GLU LYS THR TRP ASN VAL GLY SER SER ASN ARG ASN LYS ALA GLU ASN LEU LEU ARG GLY LYS ARG ASP GLY THR PHE LEU VAL ARG GLU SER SER LYS GLN GLY CYS TYR
ALA CYS SER VAL VAL VAL ASP GLY GLU VAL LYS HIS CYS VAL ILE ASN LYS THR ALA THR GLY TYR GLY PHE ALA GLU PRO TYR ASN LEU TYR SER SER LEU LYS GLU LEU VAL LEU HIS TYR GLN HIS THR SER LEU VAL GLN HIS ASN ASP SER LEU ASN VAL THR LEU ALA TYR PRO
VAL TYR ALA GLN GLN ARG ARG

4 Experimental information (i)

Property	Value	Source
EM reconstruction method	SINGLE PARTICLE	Depositor
Imposed symmetry	POINT, Not provided	
Number of particles used	506412	Depositor
Resolution determination method	DIFFRACTION PATTERN/LAYERLINES	Depositor
CTF correction method	PHASE FLIPPING AND AMPLITUDE CORRECTION	Depositor
Microscope	FEI TITAN KRIOS	Depositor
Voltage (kV)	300	Depositor
Electron dose ($e^-/Å^2$)	70	Depositor
Minimum defocus (nm)	1500	Depositor
Maximum defocus (nm)	2500	Depositor
Magnification	Not provided	
Image detector	GATAN K3 (6k x 4k)	Depositor
Maximum map value	18.853	Depositor
Minimum map value	-9.282	Depositor
Average map value	-0.000	Depositor
Map value standard deviation	1.000	Depositor
Recommended contour level	1.0	Depositor
Map size (Å)	107.00001, 133.75, 117.700005	wwPDB
Map dimensions	220, 250, 200	wwPDB
Map angles (°)	90.0, 90.0, 90.0	wwPDB
Pixel spacing (Å)	0.535, 0.535, 0.535	Depositor

5 Model quality

5.1 Standard geometry

The Z score for a bond length (or angle) is the number of standard deviations the observed value is removed from the expected value. A bond length (or angle) with $|Z| > 5$ is considered an outlier worth inspection. RMSZ is the root-mean-square of all Z scores of the bond lengths (or angles).

Mol	Chain	Bond lengths		Bond angles	
		RMSZ	$\#\|Z\| > 5$	RMSZ	$\#\|Z\| > 5$
1	A	0.25	0/8412	0.51	0/11358
2	B	0.26	0/2380	0.53	1/3187 (0.0%)
All	All	0.26	0/10792	0.51	1/14545 (0.0%)

There are no bond length outliers.

All (1) bond angle outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(^{o})	Ideal(^{o})
2	B	322	MET	CA-CB-CG	5.22	122.17	113.30

There are no chirality outliers.

There are no planarity outliers.

5.2 Too-close contacts

In the following table, the Non-H and H(model) columns list the number of non-hydrogen atoms and hydrogen atoms in the chain respectively. The H(added) column lists the number of hydrogen atoms added and optimized by MolProbity. The Clashes column lists the number of clashes within the asymmetric unit, whereas Symm-Clashes lists symmetry-related clashes.

Mol	Chain	Non-H	H(model)	H(added)	Clashes	Symm-Clashes
1	A	8233	0	8257	112	0
2	B	2341	0	2312	47	0
3	A	20	0	0	0	0
3	B	1	0	0	0	0
All	All	10595	0	10569	147	0

The all-atom clashscore is defined as the number of clashes found per 1000 atoms (including hydrogen atoms). The all-atom clashscore for this structure is 7.

The worst 5 of 147 close contacts within the same asymmetric unit are listed below, sorted by their clash magnitude.

Atom-1	Atom-2	Interatomic distance (Å)	Clash overlap (Å)
2:B:319:ASN:HD22	2:B:319:ASN:N	1.54	1.04
2:B:473:THR:HG23	2:B:552:GLN:HE21	1.46	0.81
1:A:26:LEU:HB2	1:A:30:MET:HB3	1.68	0.76
2:B:319:ASN:N	2:B:319:ASN:ND2	2.29	0.74
1:A:542:GLU:HG3	2:B:340:ARG:HH11	1.53	0.73

There are no symmetry-related clashes.

5.3 Torsion angles (i)

5.3.1 Protein backbone (i)

In the following table, the Percentiles column shows the percent Ramachandran outliers of the chain as a percentile score with respect to all PDB entries followed by that with respect to all EM entries.

The Analysed column shows the number of residues for which the backbone conformation was analysed, and the total number of residues.

Mol	Chain	Analysed	Favoured	Allowed	Outliers	Percentiles	
1	A	996/1096 (91%)	952 (96%)	44 (4%)	0	100	100
2	B	272/723 (38%)	262 (96%)	10 (4%)	0	100	100
All	All	1268/1819 (70%)	1214 (96%)	54 (4%)	0	100	100

There are no Ramachandran outliers to report.

5.3.2 Protein sidechains (i)

In the following table, the Percentiles column shows the percent sidechain outliers of the chain as a percentile score with respect to all PDB entries followed by that with respect to all EM entries.

The Analysed column shows the number of residues for which the sidechain conformation was analysed, and the total number of residues.

Mol	Chain	Analysed	Rotameric	Outliers	Percentiles	
1	A	921/999 (92%)	866 (94%)	55 (6%)	19	30
2	B	256/653 (39%)	232 (91%)	24 (9%)	8	12
All	All	1177/1652 (71%)	1098 (93%)	79 (7%)	20	25

5 of 79 residues with a non-rotameric sidechain are listed below:

Mol	Chain	Res	Type
2	B	330	ASP
2	B	474	SER
2	B	352	ASP
2	B	432	GLN
2	B	529	ASP

Sometimes sidechains can be flipped to improve hydrogen bonding and reduce clashes. All (2) such sidechains are listed below:

Mol	Chain	Res	Type
1	A	1047	HIS
2	B	552	GLN

5.3.3 RNA

There are no RNA molecules in this entry.

5.4 Non-standard residues in protein, DNA, RNA chains

There are no non-standard protein/DNA/RNA residues in this entry.

5.5 Carbohydrates

There are no monosaccharides in this entry.

5.6 Ligand geometry

There are no ligands in this entry.

5.7 Other polymers

There are no such residues in this entry.

5.8 Polymer linkage issues

There are no chain breaks in this entry.

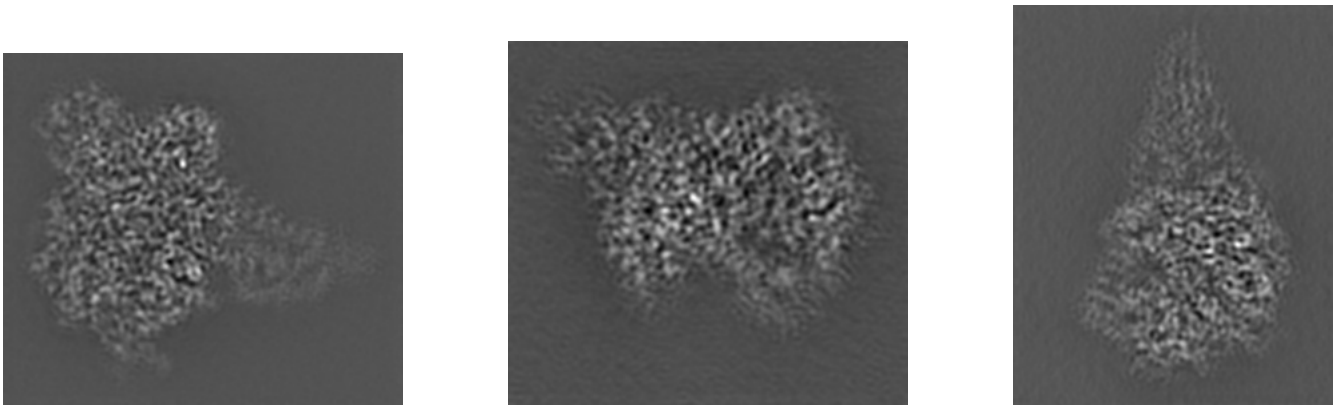
6 Map visualisation

This section contains visualisations of the EMDB entry EMD-27327. These allow visual inspection of the internal detail of the map and identification of artifacts.

Images derived from a raw map, generated by summing the deposited half-maps, are presented below the corresponding image components of the primary map to allow further visual inspection and comparison with those of the primary map.

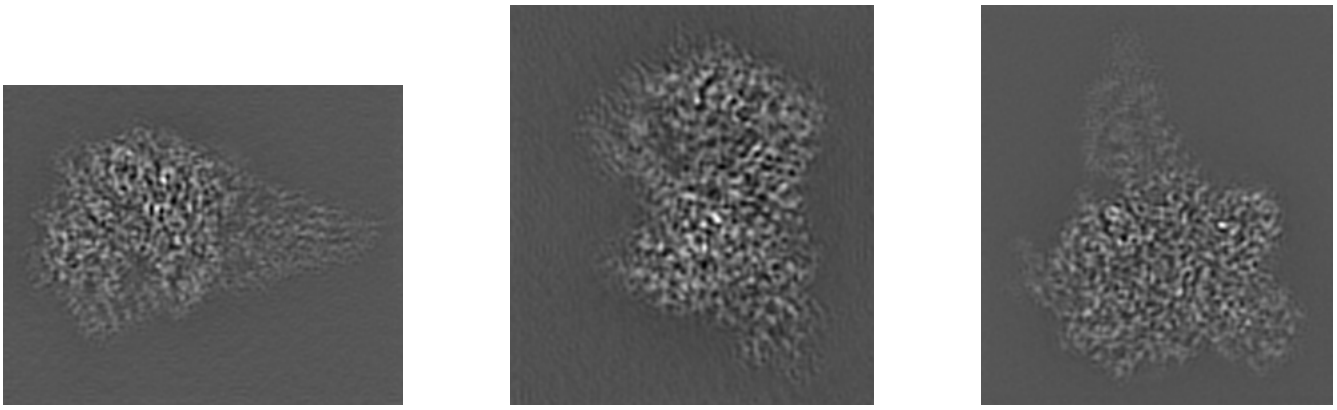
6.1 Orthogonal projections

6.1.1 Primary map



X Y Z

6.1.2 Raw map

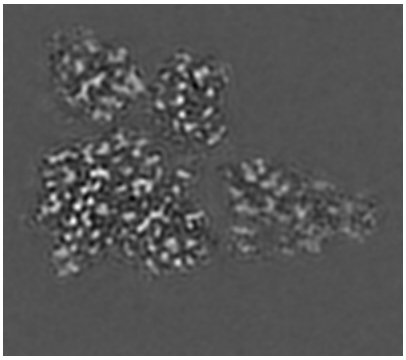


X Y Z

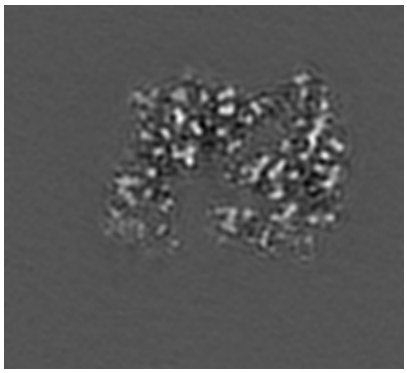
The images above show the map projected in three orthogonal directions.

6.2 Central slices (i)

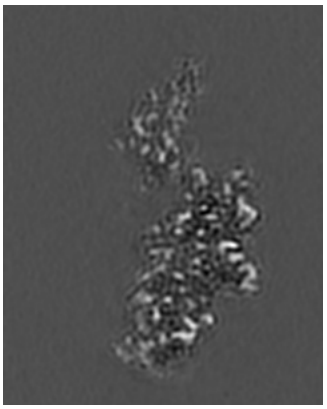
6.2.1 Primary map



X Index: 100

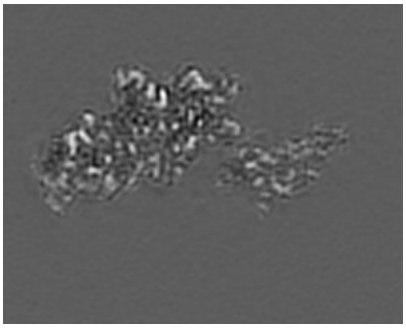


Y Index: 125

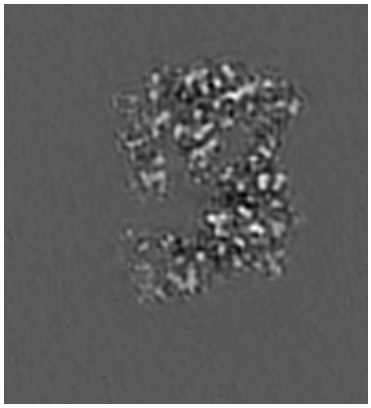


Z Index: 110

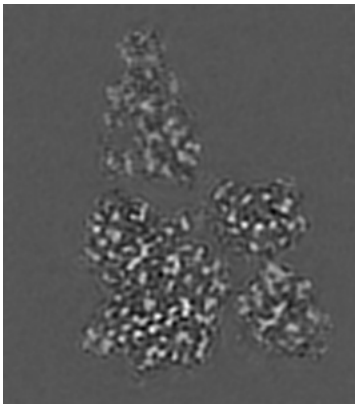
6.2.2 Raw map



X Index: 110



Y Index: 125

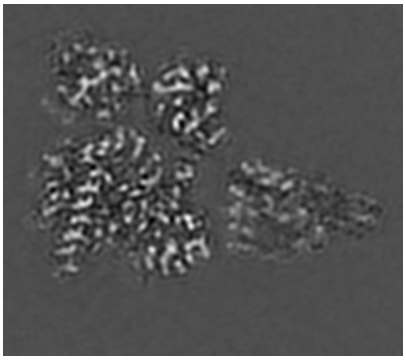


Z Index: 100

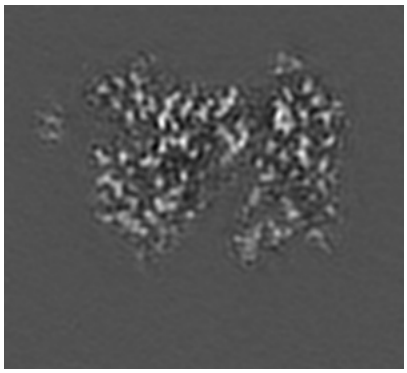
The images above show central slices of the map in three orthogonal directions.

6.3 Largest variance slices (i)

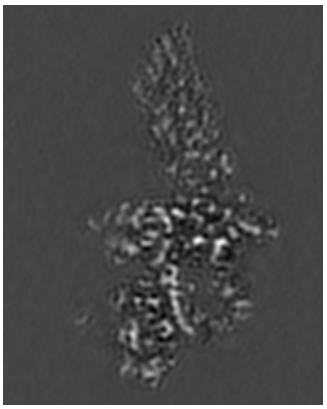
6.3.1 Primary map



X Index: 102

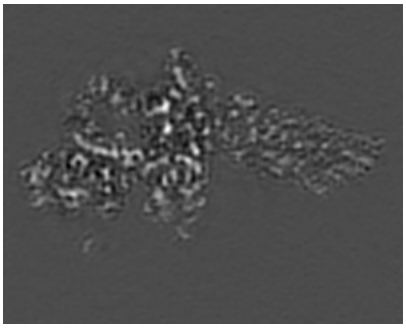


Y Index: 101

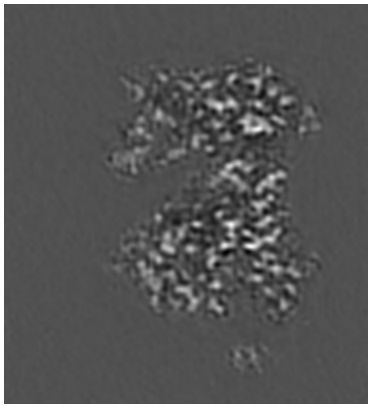


Z Index: 86

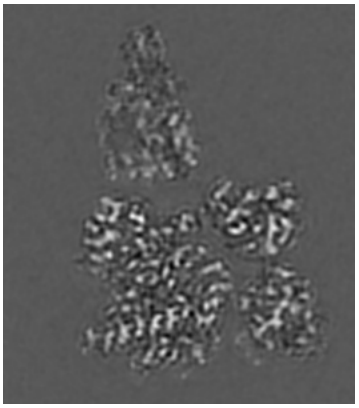
6.3.2 Raw map



X Index: 86



Y Index: 101

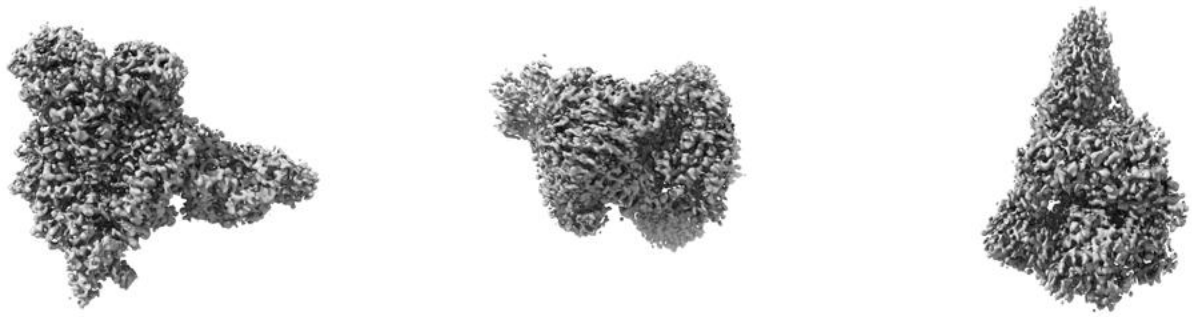


Z Index: 102

The images above show the largest variance slices of the map in three orthogonal directions.

6.4 Orthogonal surface views (i)

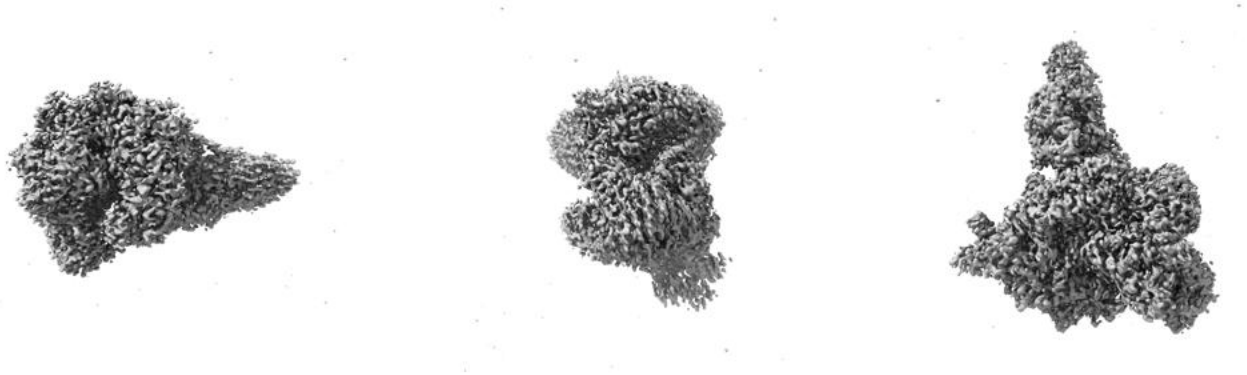
6.4.1 Primary map



X Y Z

The images above show the 3D surface view of the map at the recommended contour level 1.0. These images, in conjunction with the slice images, may facilitate assessment of whether an appropriate contour level has been provided.

6.4.2 Raw map



X Y Z

These images show the 3D surface of the raw map. The raw map's contour level was selected so that its surface encloses the same volume as the primary map does at its recommended contour level.

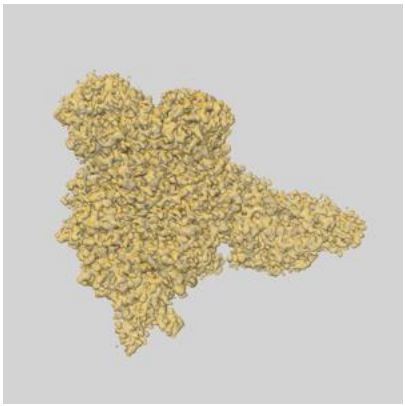
6.5 Mask visualisation (i)

This section shows the 3D surface view of the primary map at 50% transparency overlaid with the specified mask at 0% transparency

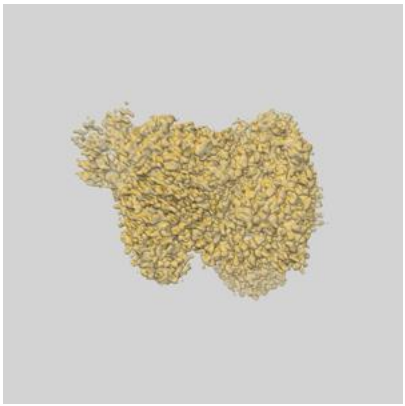
A mask typically either:

- Encompasses the whole structure
- Separates out a domain, a functional unit, a monomer or an area of interest from a larger structure

6.5.1 emd_27327_msk_1.map (i)



X



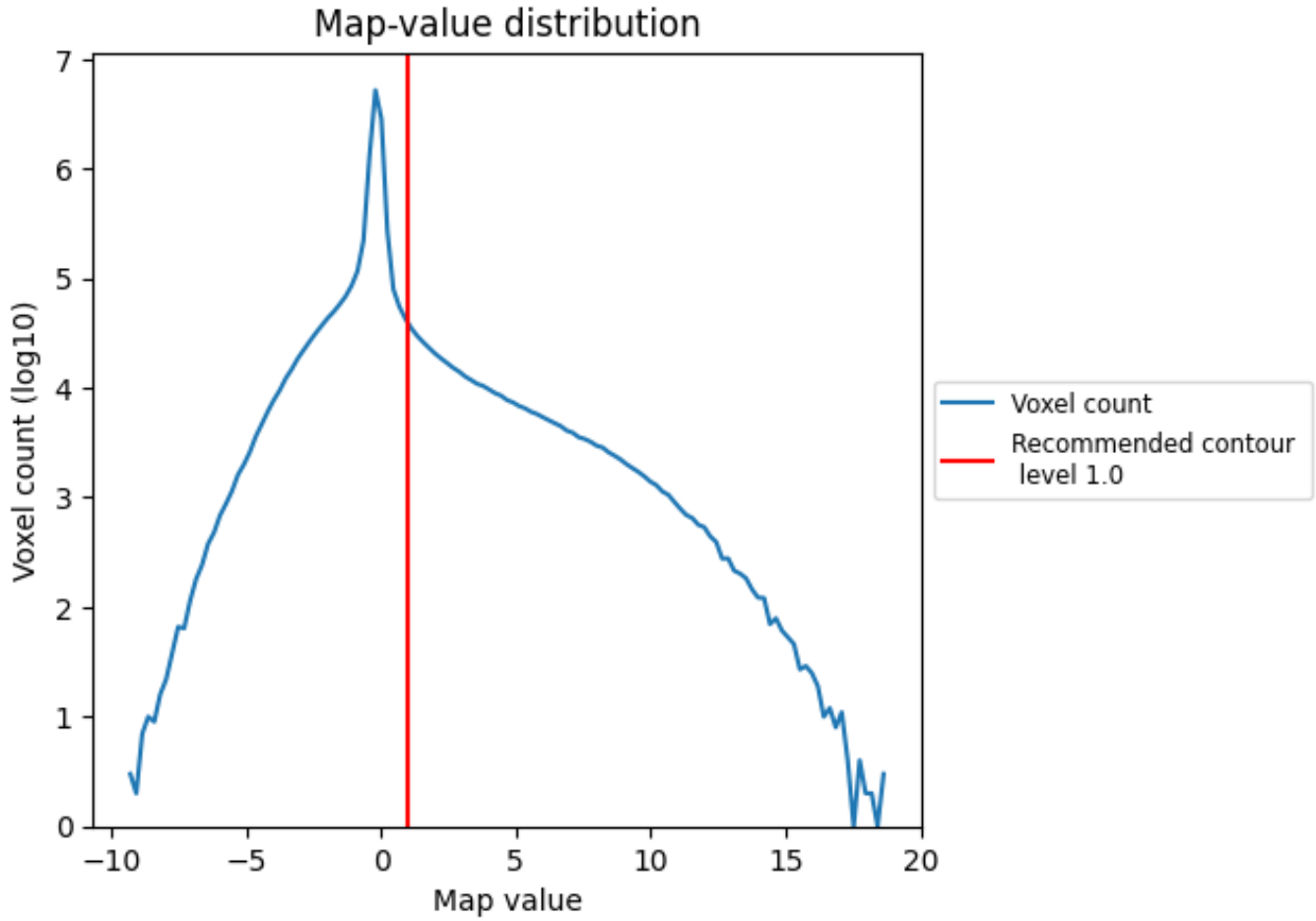
Y

Z

7 Map analysis

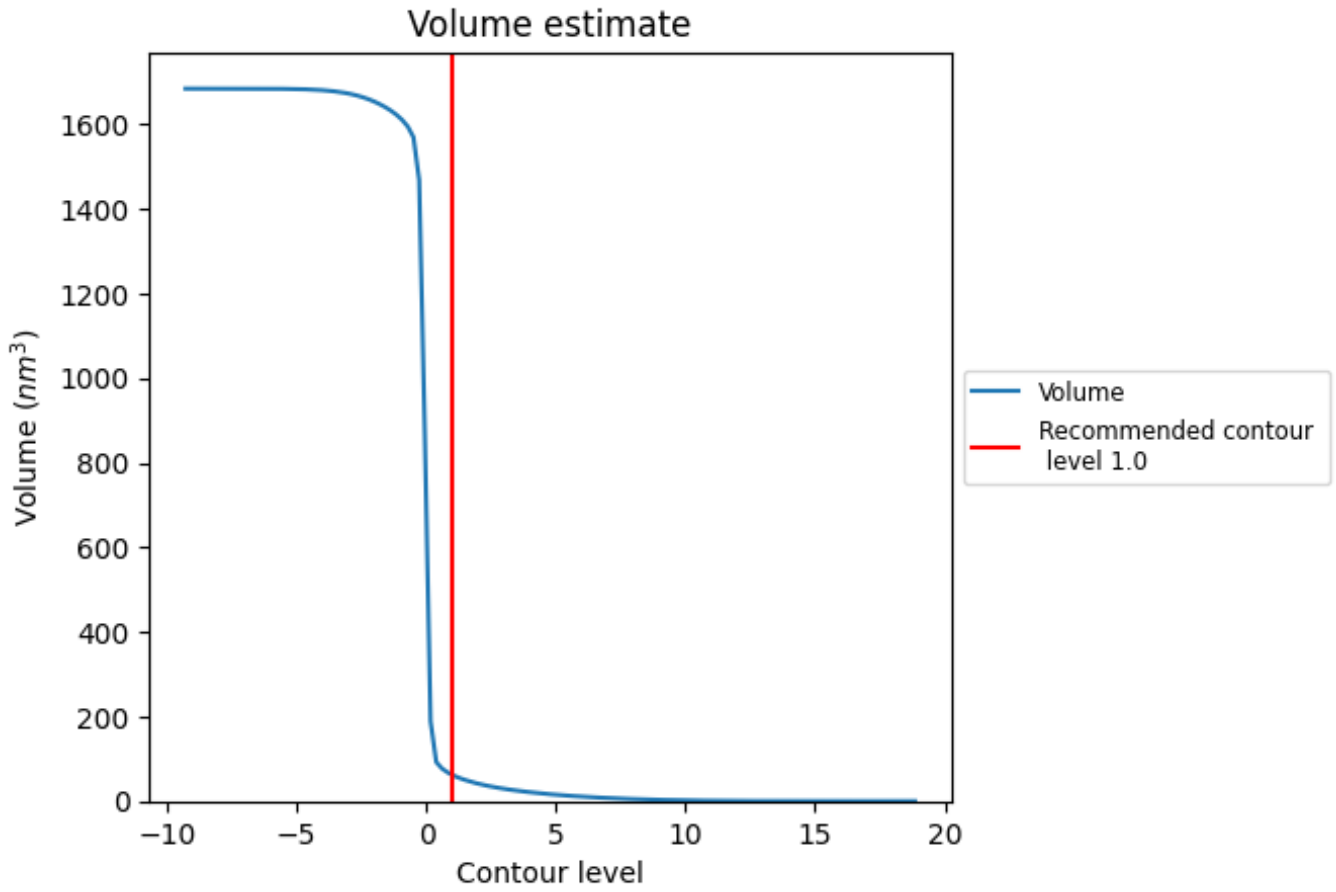
This section contains the results of statistical analysis of the map.

7.1 Map-value distribution



The map-value distribution is plotted in 128 intervals along the x-axis. The y-axis is logarithmic. A spike in this graph at zero usually indicates that the volume has been masked.

7.2 Volume estimate (i)



The volume at the recommended contour level is 62 nm^3; this corresponds to an approximate mass of 56 kDa.

The volume estimate graph shows how the enclosed volume varies with the contour level. The recommended contour level is shown as a vertical line and the intersection between the line and the curve gives the volume of the enclosed surface at the given level.

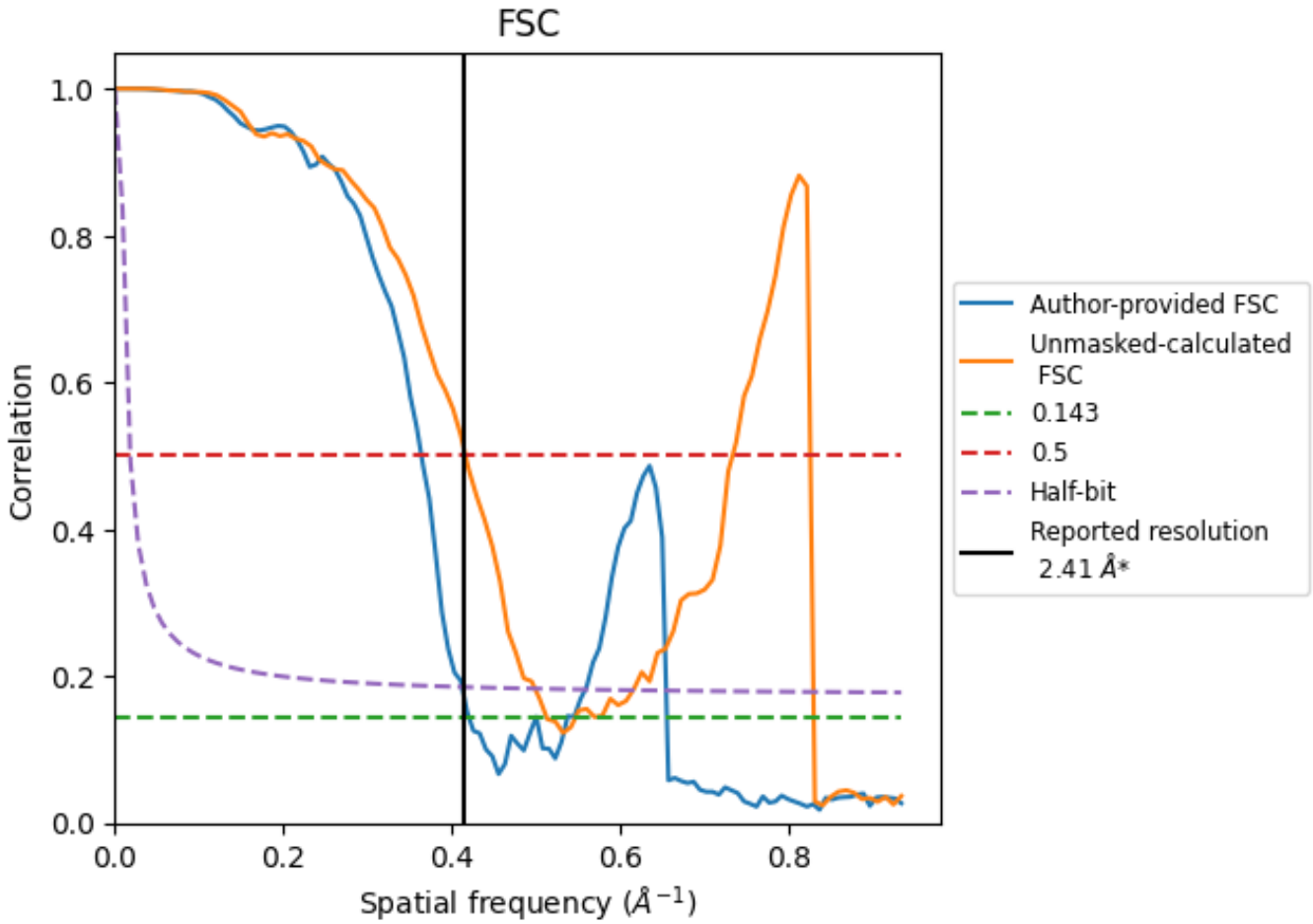
7.3 Rotationally averaged power spectrum (i)

This section was not generated. The rotationally averaged power spectrum is only generated for cubic maps.

8 Fourier-Shell correlation

Fourier-Shell Correlation (FSC) is the most commonly used method to estimate the resolution of single-particle and subtomogram-averaged maps. The shape of the curve depends on the imposed symmetry, mask and whether or not the two 3D reconstructions used were processed from a common reference. The reported resolution is shown as a black line. A curve is displayed for the half-bit criterion in addition to lines showing the 0.143 gold standard cut-off and 0.5 cut-off.

8.1 FSC



*Reported resolution corresponds to spatial frequency of 0.415 Å^{-1}

8.2 Resolution estimates (i)

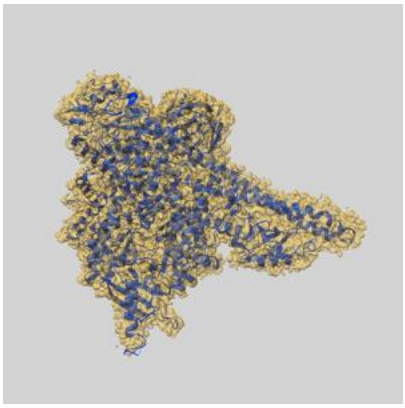
Resolution estimate (Å)	Estimation criterion (FSC cut-off)		
	0.143	0.5	Half-bit
Reported by author	-	-	-
Author-provided FSC curve	2.38	2.74	2.42
Unmasked-calculated*	1.95	2.40	2.00

*Resolution estimate based on FSC curve calculated by comparison of deposited half-maps.

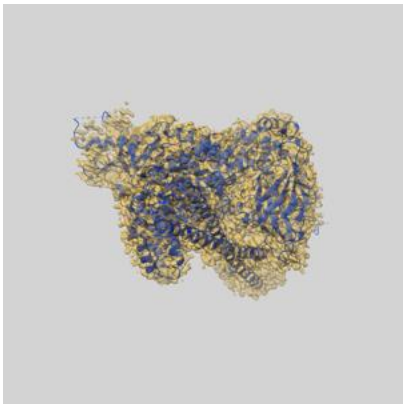
9 Map-model fit

This section contains information regarding the fit between EMDB map EMD-27327 and PDB model 8DCP. Per-residue inclusion information can be found in section 3 on page 5.

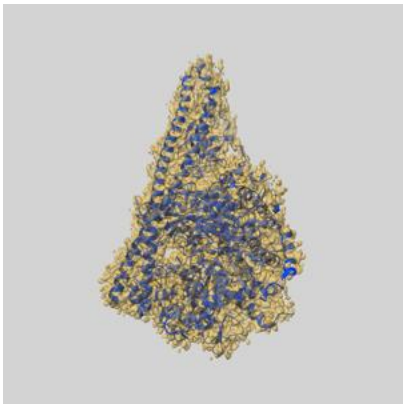
9.1 Map-model overlay



X



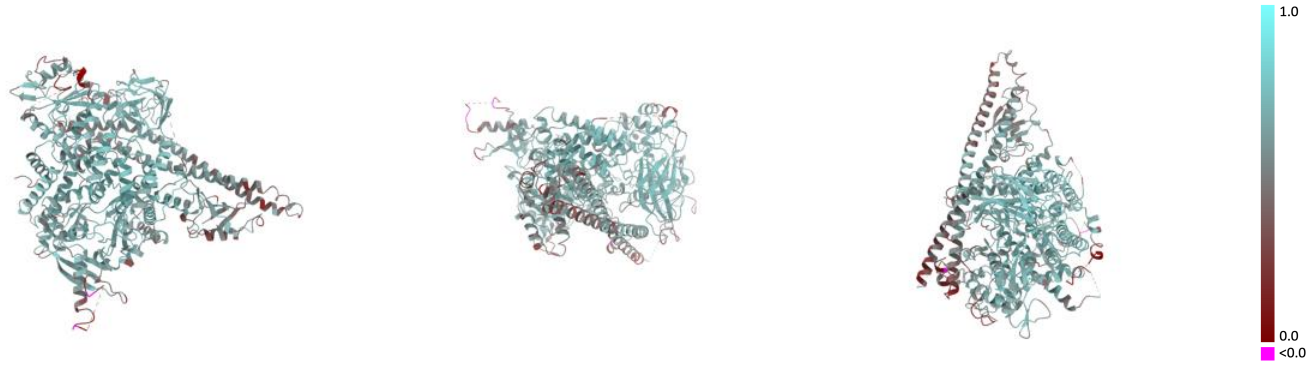
Y



Z

The images above show the 3D surface view of the map at the recommended contour level 1.0 at 50% transparency in yellow overlaid with a ribbon representation of the model coloured in blue. These images allow for the visual assessment of the quality of fit between the atomic model and the map.

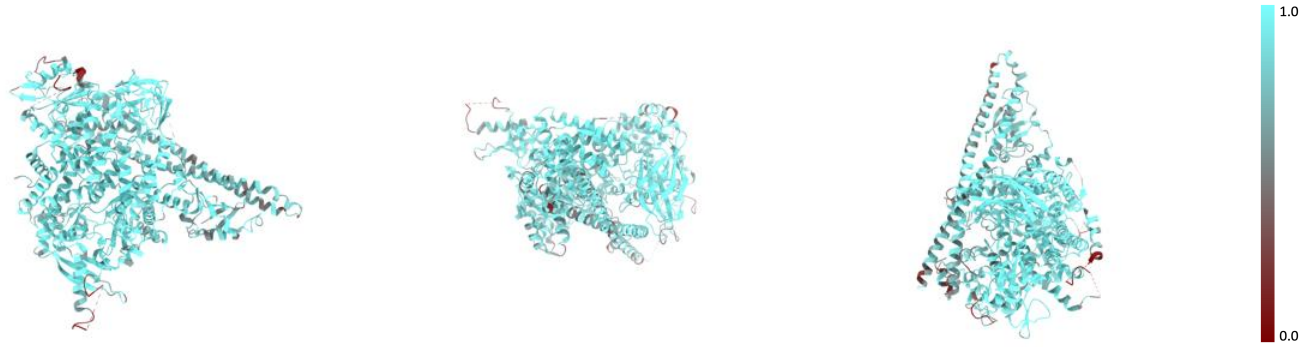
9.2 Q-score mapped to coordinate model



X Y Z

The images above show the model with each residue coloured according its Q-score. This shows their resolvability in the map with higher Q-score values reflecting better resolvability. Please note: Q-score is calculating the resolvability of atoms, and thus high values are only expected at resolutions at which atoms can be resolved. Low Q-score values may therefore be expected for many entries.

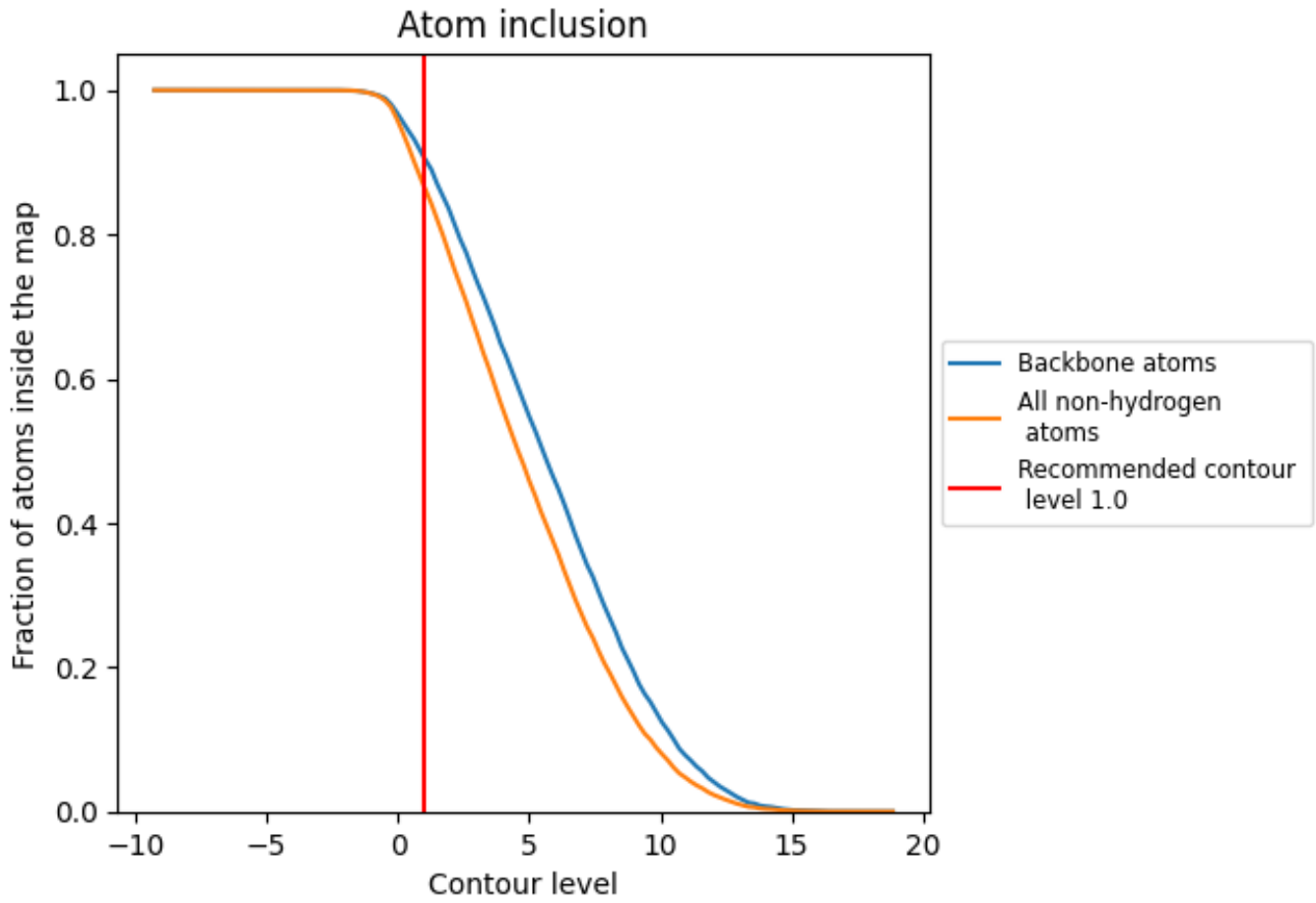
9.3 Atom inclusion mapped to coordinate model



X Y Z

The images above show the model with each residue coloured according to its atom inclusion. This shows to what extent they are inside the map at the recommended contour level (1.0).

9.4 Atom inclusion (i)



At the recommended contour level, 91% of all backbone atoms, 87% of all non-hydrogen atoms, are inside the map.

9.5 Map-model fit summary (i)

The table lists the average atom inclusion at the recommended contour level (1.0) and Q-score for the entire model and for each chain.

Chain	Atom inclusion	Q-score
All	0.8653	0.5840
A	0.8875	0.6080
B	0.7923	0.5020