wwPDB EM Validation Summary Report

Nov 4, 2024 – 06:15 AM JST

PDB ID	:	7W53
EMDB ID	:	EMD-32313
Title	:	Cryo-EM structure of the neuromedin U-bound neuromedin U receptor 1-Gq protein complex
Authors	:	You, C.; Xu, H.E.; Jiang, Y.
Deposited on	:	2021-11-29
Resolution	:	3.20 Å(reported)

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:

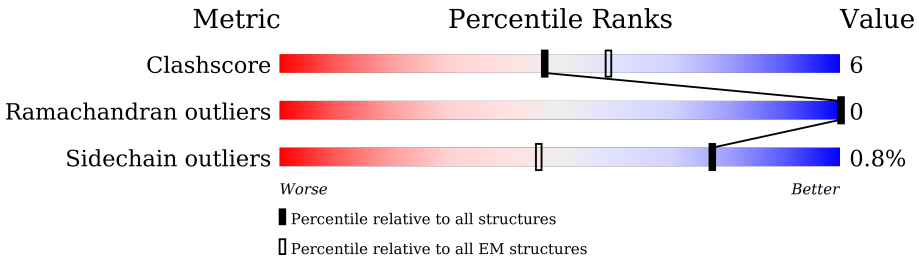
EMDB validation analysis	:	0.0.1.dev113
MolProbity	:	4.02b-467
Percentile statistics	:	20231227.v01 (using entries in the PDB archive December 27th 2023)
MapQ	:	1.9.13
Ideal geometry (proteins)	:	Engh & Huber (2001)
Ideal geometry (DNA, RNA)	:	Parkinson et al. (1996)
Validation Pipeline (wwPDB-VP)	:	2.39

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 3.20 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	210492	15764
Ramachandran outliers	207382	16835
Sidechain outliers	206894	16415

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	361	11% 50% 7% 42%
2	B	371	9% 73% 18% 9%
3	C	26	12% 19% 12% 69%
4	E	249	10% 78% 15% 7%
5	G	70	30% 71% 10% 19%
6	R	755	14% 31% 65%

2 Entry composition (i)

There are 6 unique types of molecules in this entry. The entry contains 17354 atoms, of which 8644 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 Guanine nucleotide-binding protein G(q) subunit alpha.

Mol	Chain	Residues	Atoms						AltConf	Trace
			Total	C	H	N	O	S		
1	A	210	3443	1095	1709	309	323	7	0	0

- Molecule 2 is a protein called Guanine nucleotide-binding protein G(I)/G(S)/G(T) subunit beta-1.

Mol	Chain	Residues	Atoms						AltConf	Trace
			Total	C	H	N	O	S		
2	B	338	5109	1604	2509	467	508	21	0	0

There are 32 discrepancies between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	Comment	Reference
B	1	MET	-	initiating methionine	UNP P62873
B	2	GLY	-	expression tag	UNP P62873
B	3	SER	-	expression tag	UNP P62873
B	4	LEU	-	expression tag	UNP P62873
B	5	LEU	-	expression tag	UNP P62873
B	6	GLN	-	expression tag	UNP P62873
B	346	GLY	-	expression tag	UNP P62873
B	347	SER	-	expression tag	UNP P62873
B	348	SER	-	expression tag	UNP P62873
B	349	GLY	-	expression tag	UNP P62873
B	350	GLY	-	expression tag	UNP P62873
B	351	GLY	-	expression tag	UNP P62873
B	352	GLY	-	expression tag	UNP P62873
B	353	SER	-	expression tag	UNP P62873
B	354	GLY	-	expression tag	UNP P62873
B	355	GLY	-	expression tag	UNP P62873
B	356	GLY	-	expression tag	UNP P62873
B	357	GLY	-	expression tag	UNP P62873
B	358	SER	-	expression tag	UNP P62873
B	359	SER	-	expression tag	UNP P62873

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Chain	Residue	Modelled	Actual	Comment	Reference
B	360	GLY	-	expression tag	UNP P62873
B	361	VAL	-	expression tag	UNP P62873
B	362	SER	-	expression tag	UNP P62873
B	363	GLY	-	expression tag	UNP P62873
B	364	TRP	-	expression tag	UNP P62873
B	365	ARG	-	expression tag	UNP P62873
B	366	LEU	-	expression tag	UNP P62873
B	367	PHE	-	expression tag	UNP P62873
B	368	LYS	-	expression tag	UNP P62873
B	369	LYS	-	expression tag	UNP P62873
B	370	ILE	-	expression tag	UNP P62873
B	371	SER	-	expression tag	UNP P62873

- Molecule 3 is a protein called Neuromedin-U-25.

Mol	Chain	Residues	Atoms					AltConf	Trace
3	C	8	Total	C	H	N	O	0	1
			136	45	68	15	8		

There is a discrepancy between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	Comment	Reference
C	10	GLN	-	expression tag	UNP P48645

- Molecule 4 is a protein called scFv16.

Mol	Chain	Residues	Atoms						AltConf	Trace
4	E	232	Total	C	H	N	O	S	0	0
			3504	1131	1722	295	346	10		

- Molecule 5 is a protein called Guanine nucleotide-binding protein G(I)/G(S)/G(O) subunit gamma-2.

Mol	Chain	Residues	Atoms						AltConf	Trace
5	G	57	Total	C	H	N	O	S	0	0
			885	273	449	77	83	3		

- Molecule 6 is a protein called Neuromedin-U receptor 1.

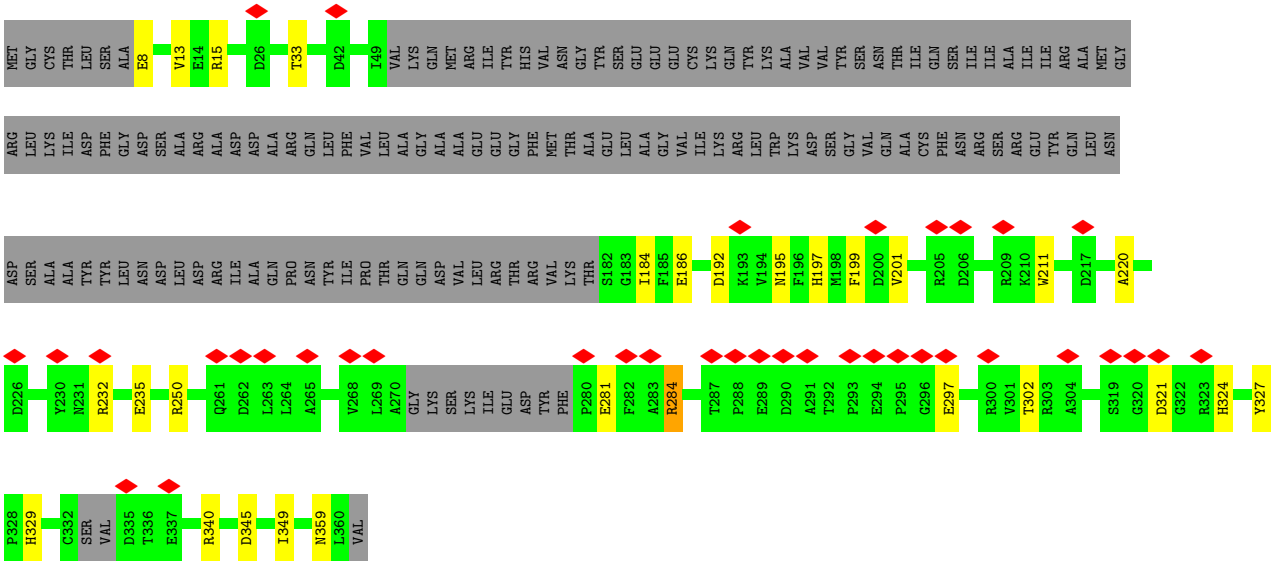
Mol	Chain	Residues	Atoms						AltConf	Trace
6	R	264	Total	C	H	N	O	S	0	0
			4277	1373	2187	361	335	21		

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.

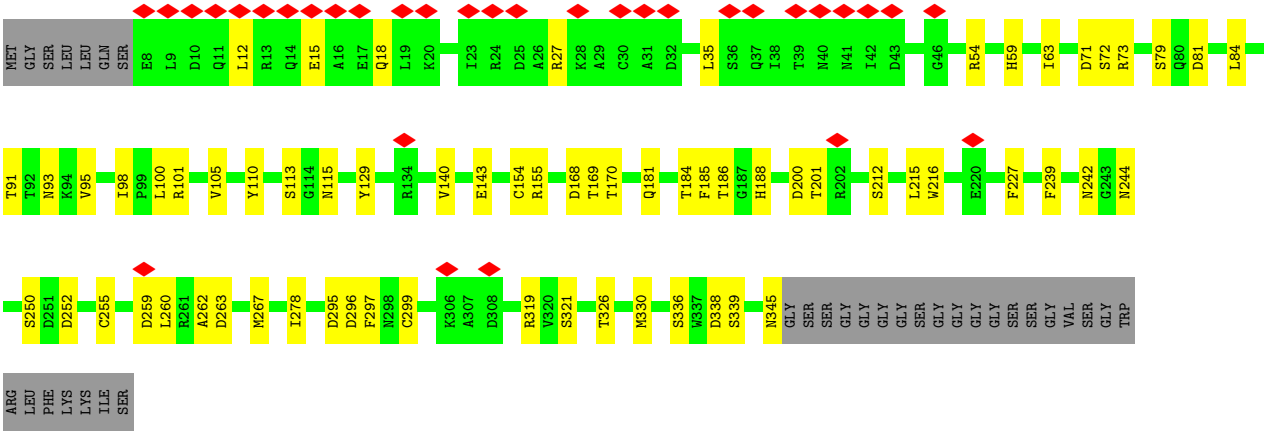
• Molecule 1: Guanine nucleotide-binding protein G(q) subunit alpha

Chain A:



• Molecule 2: Guanine nucleotide-binding protein G(I)/G(S)/G(T) subunit beta-1

Chain B:

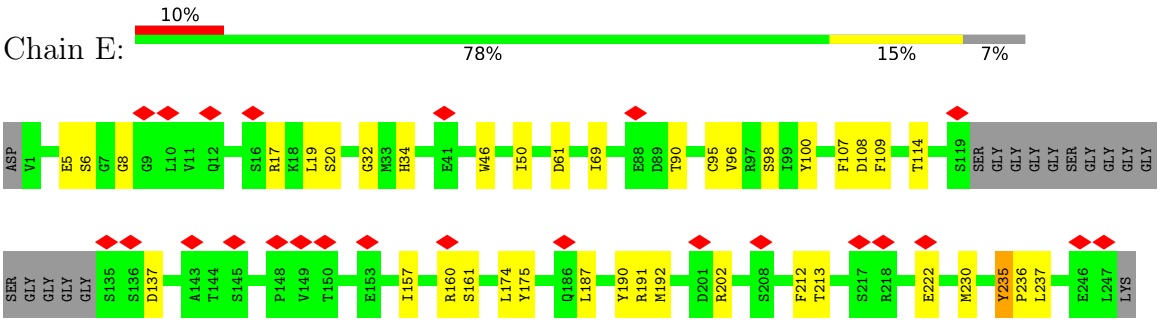


• Molecule 3: Neuromedin-U-25

Chain C:

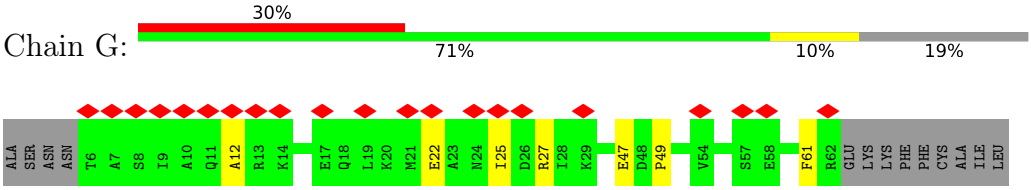
• Molecule 4: scFv16

Chain E:



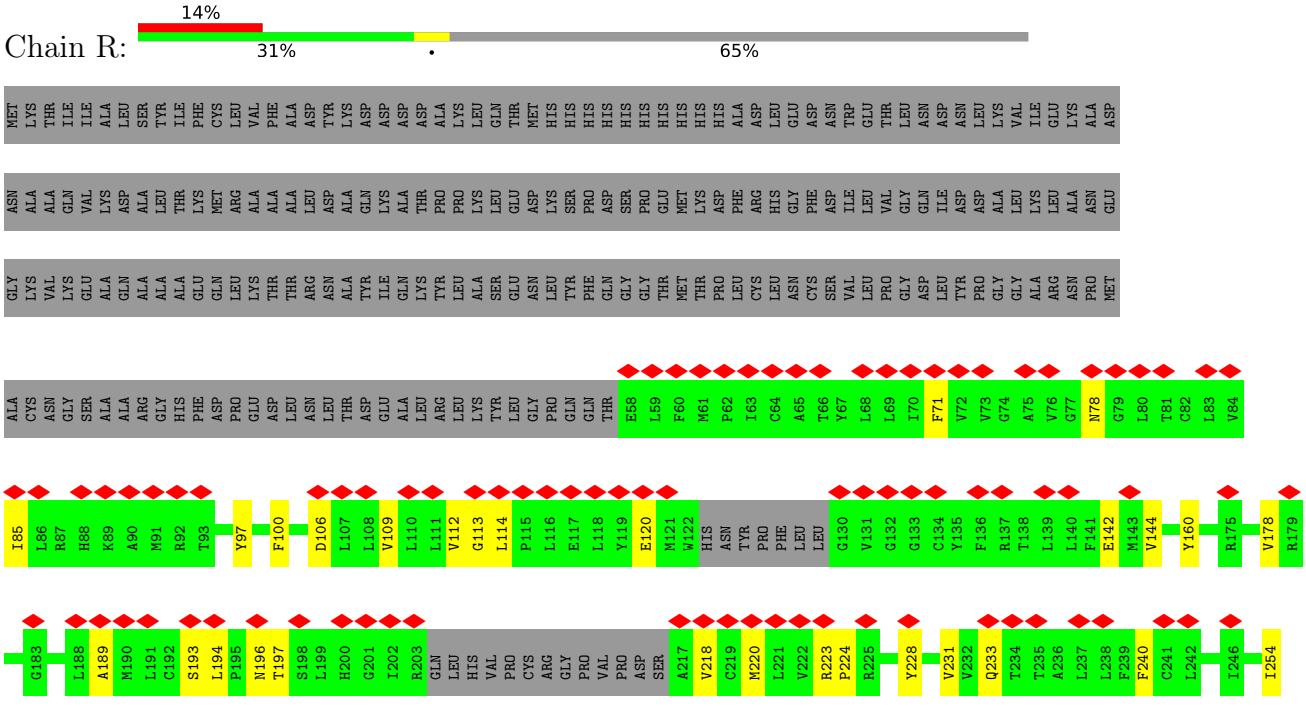
• Molecule 5: Guanine nucleotide-binding protein G(I)/G(S)/G(O) subunit gamma-2

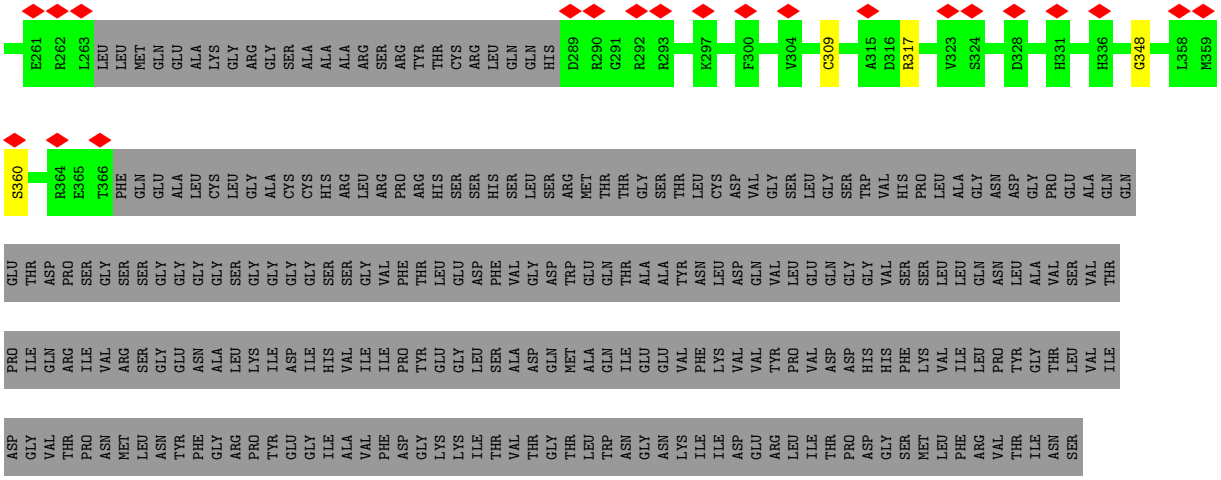
Chain G:



• Molecule 6: Neuromedin-U receptor 1

Chain R:




E261 R262 L263 LEU LEU MET GLN GLU ALA LYS GLY ARG GLY SER ALA ALA ALA ARG SER ARG TYR THR CYS ARG LEU GLN GLN HIS D289 R290 G291 R292 R293 K297 F300 V304 C309 A315 D316 R317 V323 S324 D328 H331 H336 G348 L358 M359
S360 R364 E365 T366 PHE GLN GLU ALA LEU CYS LEU GLY ALA CYS CYS HIS ARG LEU ARG PRO ARG HIS SER SER HIS SER LEU SER ARG MET THR THR GLY SER THR LEU CYS ASP VAL GLY SER LEU GLY SER TRP VAL HIS PRO LEU ALA GLY ASN ASP GLY PRO GLU ALA GLN GLN
GLU THR ASP PRO SER GLY SER SER GLY GLY GLY GLY SER GLY GLY GLY GLY SER SER GLY VAL PHE THR LEU GLU ASP PHE VAL GLY ASP TRP GLU GLN THR ALA ALA TYR ASN LEU ASP GLN VAL LEU GLU GLN GLY GLY VAL SER SER LEU LEU GLN ASN LEU ALA VAL SER VAL THR
PRO ILE GLN ARG ILE VAL ARG SER GLY GLU ASN ALA LEU LYS ILE ASP ILE HIS VAL ILE ILE PRO TYR GLU GLY LEU SER ALA ASP GLN MET ALA GLN ILE GLU GLU VAL PHE LYS VAL VAL TYR PRO VAL ASP ASP HIS HIS PHE LYS VAL ILE LEU PRO TYR GLY THR LEU VAL ILE
ASP GLY VAL THR PRO ASN MET LEU ASN TYR PHE GLY ARG PRO TYR GLU GLY ILE ALA VAL PHE ASP GLY LYS LYS ILE THR VAL THR GLY THR LEU TRP ASN GLY ASN LYS ILE ILE ILE ASP GLU ARG LEU ILE THR PRO ASP GLY SER MET LEU PHE ARG VAL THR ILE ASN SER

4 Experimental information (i)

Property	Value	Source
EM reconstruction method	SINGLE PARTICLE	Depositor
Imposed symmetry	POINT, Not provided	
Number of particles used	312310	Depositor
Resolution determination method	FSC 0.143 CUT-OFF	Depositor
CTF correction method	NONE	Depositor
Microscope	FEI TITAN KRIOS	Depositor
Voltage (kV)	300	Depositor
Electron dose ($e^-/Å^2$)	61.8	Depositor
Minimum defocus (nm)	1100	Depositor
Maximum defocus (nm)	2200	Depositor
Magnification	Not provided	
Image detector	GATAN K3 (6k x 4k)	Depositor
Maximum map value	0.134	Depositor
Minimum map value	-0.090	Depositor
Average map value	-0.000	Depositor
Map value standard deviation	0.005	Depositor
Recommended contour level	0.024	Depositor
Map size (Å)	216.00002, 216.00002, 216.00002	wwPDB
Map dimensions	200, 200, 200	wwPDB
Map angles (°)	90.0, 90.0, 90.0	wwPDB
Pixel spacing (Å)	1.08, 1.08, 1.08	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	0.37	0/1764	0.50	0/2372
2	B	0.43	0/2647	0.57	2/3589 (0.1%)
3	C	0.43	0/70	0.59	0/93
4	E	0.42	0/1826	0.59	2/2476 (0.1%)
5	G	0.31	0/442	0.40	0/597
6	R	0.33	0/2137	0.49	0/2902
All	All	0.39	0/8886	0.53	4/12029 (0.0%)

There are no bond length outliers.

All (4) bond angle outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(^{o})	Ideal(^{o})
4	E	236	PRO	N-CA-C	-11.90	81.16	112.10
4	E	235	TYR	N-CA-C	9.08	135.51	111.00
2	B	296	ASP	N-CA-C	-5.39	96.45	111.00
2	B	296	ASP	CB-CG-OD1	5.07	122.87	118.30

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	1734	1709	1717	17	0
2	B	2600	2509	2505	40	0
3	C	68	68	67	6	0

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Mol	Chain	Non-H	H(model)	H(added)	Clashes	Symm-Clashes
4	E	1782	1722	1721	24	0
5	G	436	449	448	6	0
6	R	2090	2187	2181	21	0
All	All	8710	8644	8639	100	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 6.

The worst 5 of 100 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:79:SER:OG	2:B:81:ASP:OD1	1.77	1.01
1:A:15:ARG:NH1	2:B:95:VAL:O	1.99	0.96
3:C:10:GLN:N	6:R:142:GLU:OE2	2.06	0.88
2:B:100:LEU:HD13	2:B:105:VAL:HG11	1.61	0.82
1:A:192:ASP:OD2	1:A:340:ARG:NH2	2.15	0.79

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	202/361 (56%)	192 (95%)	10 (5%)	0	100	100
2	B	336/371 (91%)	323 (96%)	13 (4%)	0	100	100
3	C	6/26 (23%)	6 (100%)	0	0	100	100
4	E	228/249 (92%)	214 (94%)	14 (6%)	0	100	100
5	G	55/70 (79%)	54 (98%)	1 (2%)	0	100	100
6	R	256/755 (34%)	252 (98%)	4 (2%)	0	100	100
All	All	1083/1832 (59%)	1041 (96%)	42 (4%)	0	100	100

There are no Ramachandran outliers to report.

5.3.2 Protein sidechains

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	190/316 (60%)	188 (99%)	2 (1%)	70 86
2	B	281/302 (93%)	278 (99%)	3 (1%)	70 86
3	C	7/24 (29%)	7 (100%)	0	100 100
4	E	196/202 (97%)	196 (100%)	0	100 100
5	G	46/57 (81%)	46 (100%)	0	100 100
6	R	228/635 (36%)	225 (99%)	3 (1%)	65 83
All	All	948/1536 (62%)	940 (99%)	8 (1%)	77 90

5 of 8 residues with a non-rotameric sidechain are listed below:

Mol	Chain	Res	Type
6	R	240	PHE
6	R	220	MET
2	B	299	CYS
2	B	239	PHE
6	R	218	VAL

Sometimes sidechains can be flipped to improve hydrogen bonding and reduce clashes. All (1) such sidechains are listed below:

Mol	Chain	Res	Type
1	A	216	ASN

5.3.3 RNA

There are no RNA molecules in this entry.

5.4 Non-standard residues in protein, DNA, RNA chains (i)

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

5.5 Carbohydrates (i)

There are no oligosaccharides in this entry.

5.6 Ligand geometry (i)

There are no ligands in this entry.

5.7 Other polymers (i)

There are no such residues in this entry.

5.8 Polymer linkage issues (i)

There are no chain breaks in this entry.

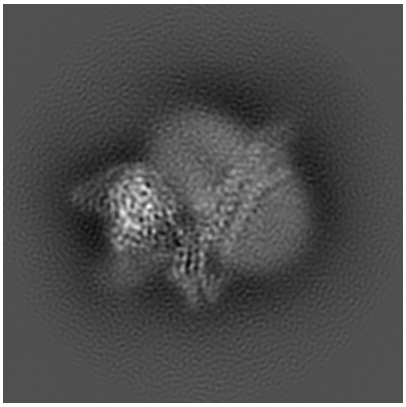
6 Map visualisation

This section contains visualisations of the EMDB entry EMD-32313. These allow visual inspection of the internal detail of the map and identification of artifacts.

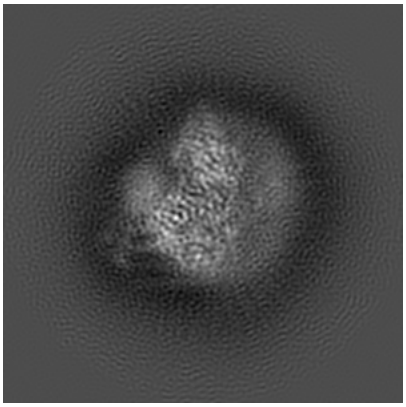
No raw map or half-maps were deposited for this entry and therefore no images, graphs, etc. pertaining to the raw map can be shown.

6.1 Orthogonal projections

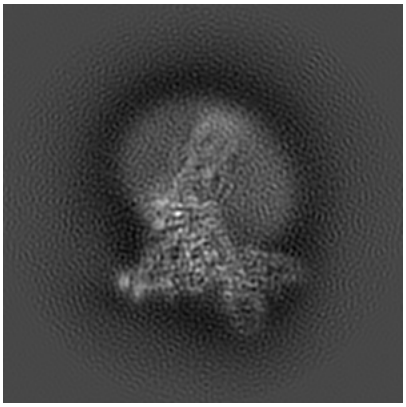
6.1.1 Primary map



X



Y

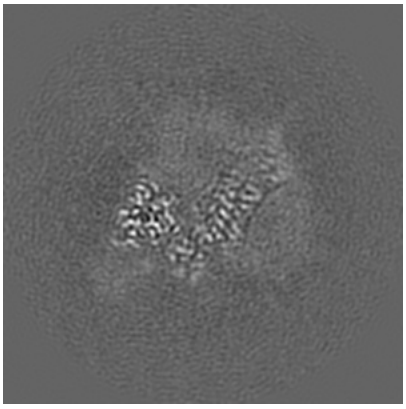


Z

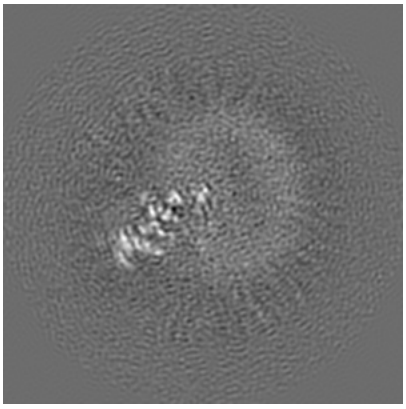
The images above show the map projected in three orthogonal directions.

6.2 Central slices

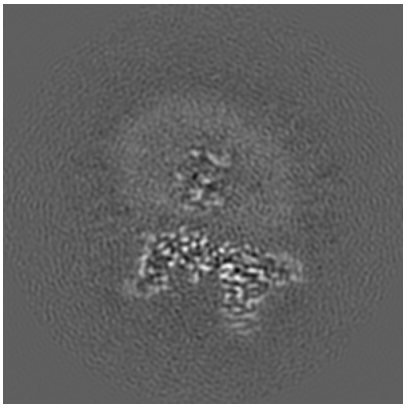
6.2.1 Primary map



X Index: 100



Y Index: 100

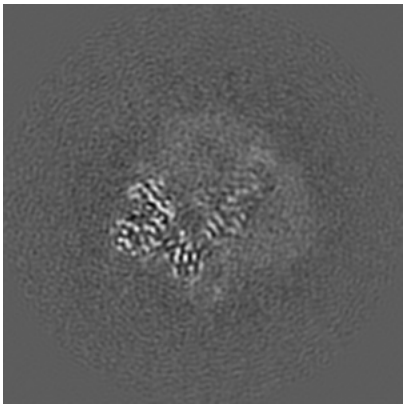


Z Index: 100

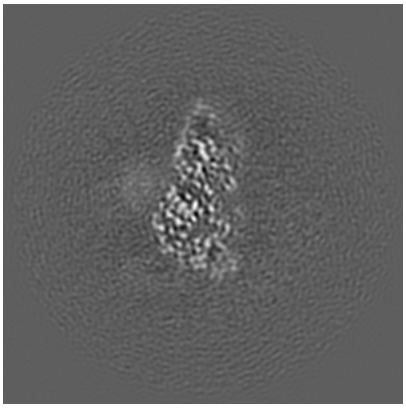
The images above show central slices of the map in three orthogonal directions.

6.3 Largest variance slices (i)

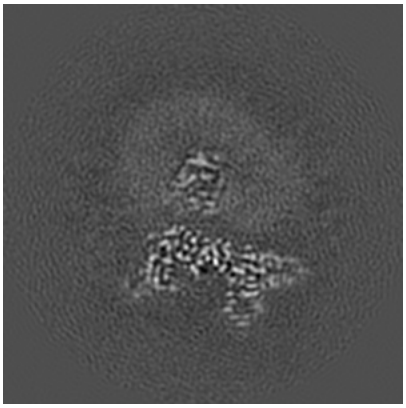
6.3.1 Primary map



X Index: 91



Y Index: 66

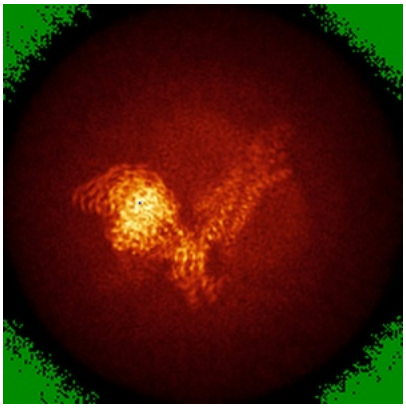


Z Index: 98

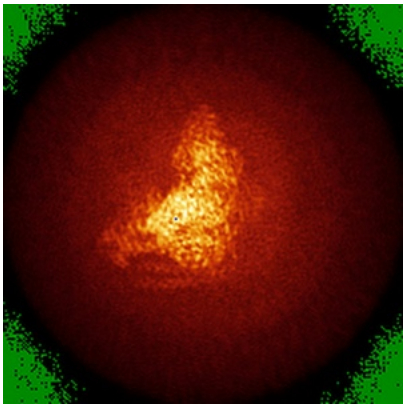
The images above show the largest variance slices of the map in three orthogonal directions.

6.4 Orthogonal standard-deviation projections (False-color) (i)

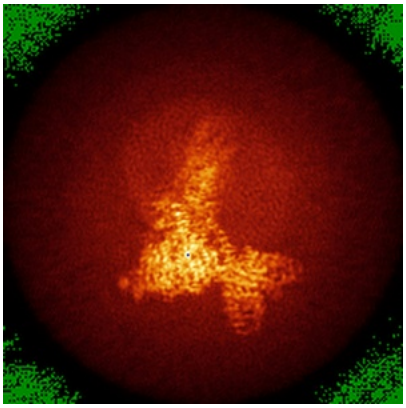
6.4.1 Primary map



X



Y



Z

The images above show the map standard deviation projections with false color in three orthogonal directions. Minimum values are shown in green, max in blue, and dark to light orange shades represent small to large values respectively.

6.5 Orthogonal surface views (i)

6.5.1 Primary map

X Y Z

The images above show the 3D surface view of the map at the recommended contour level 0.024. These images, in conjunction with the slice images, may facilitate assessment of whether an appropriate contour level has been provided.

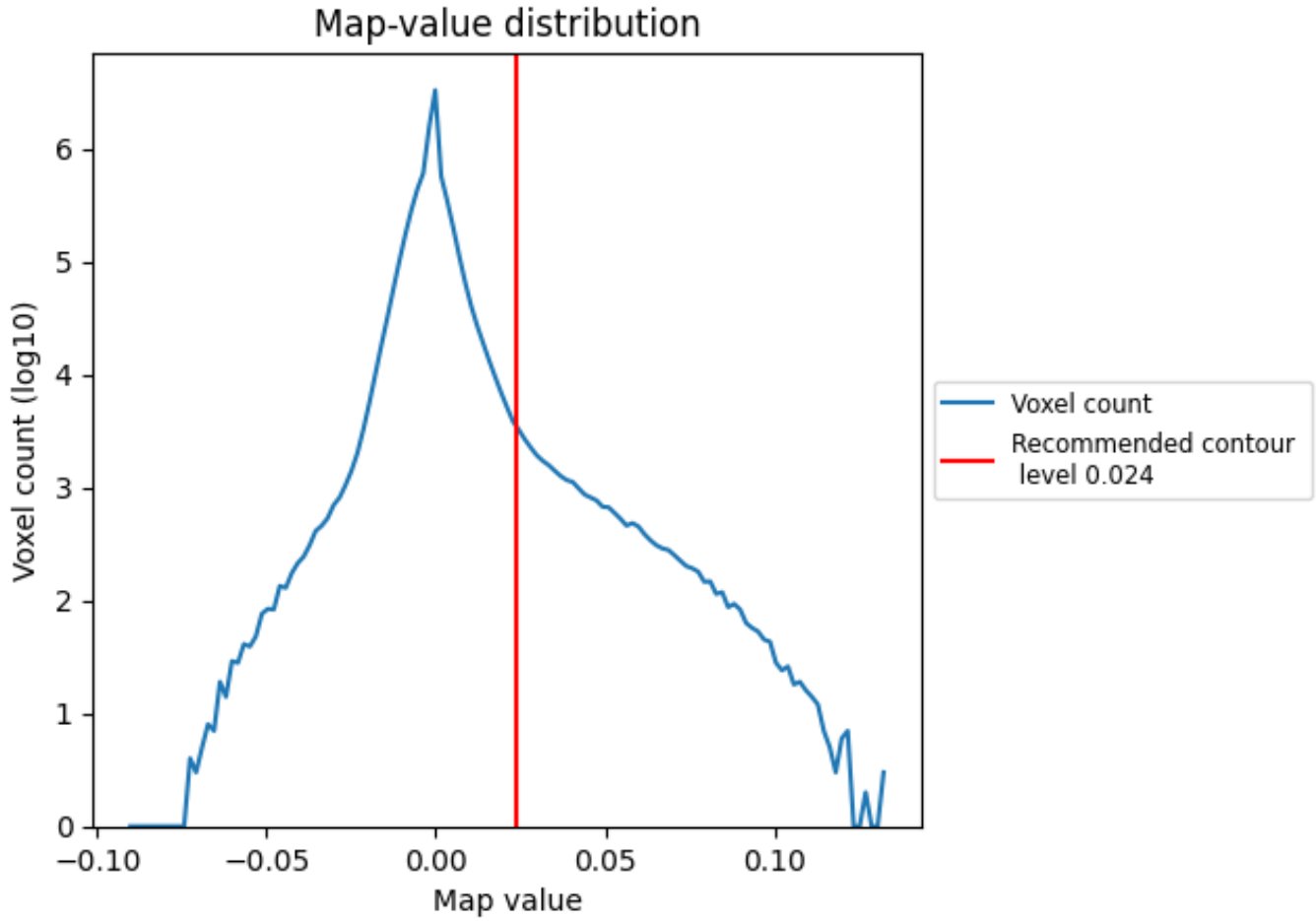
6.6 Mask visualisation (i)

This section was not generated. No masks/segmentation were deposited.

7 Map analysis

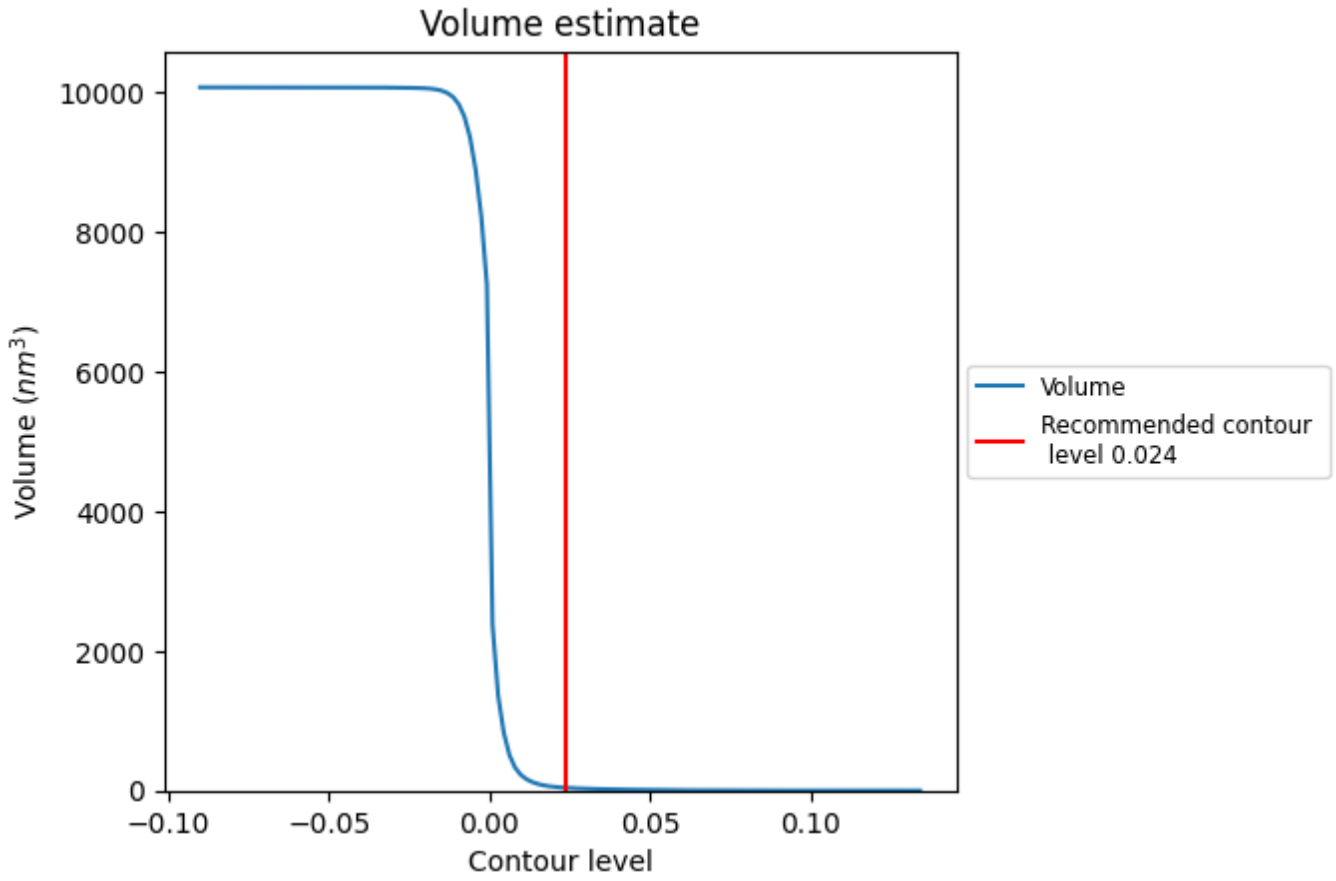
This section contains the results of statistical analysis of the map.

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.

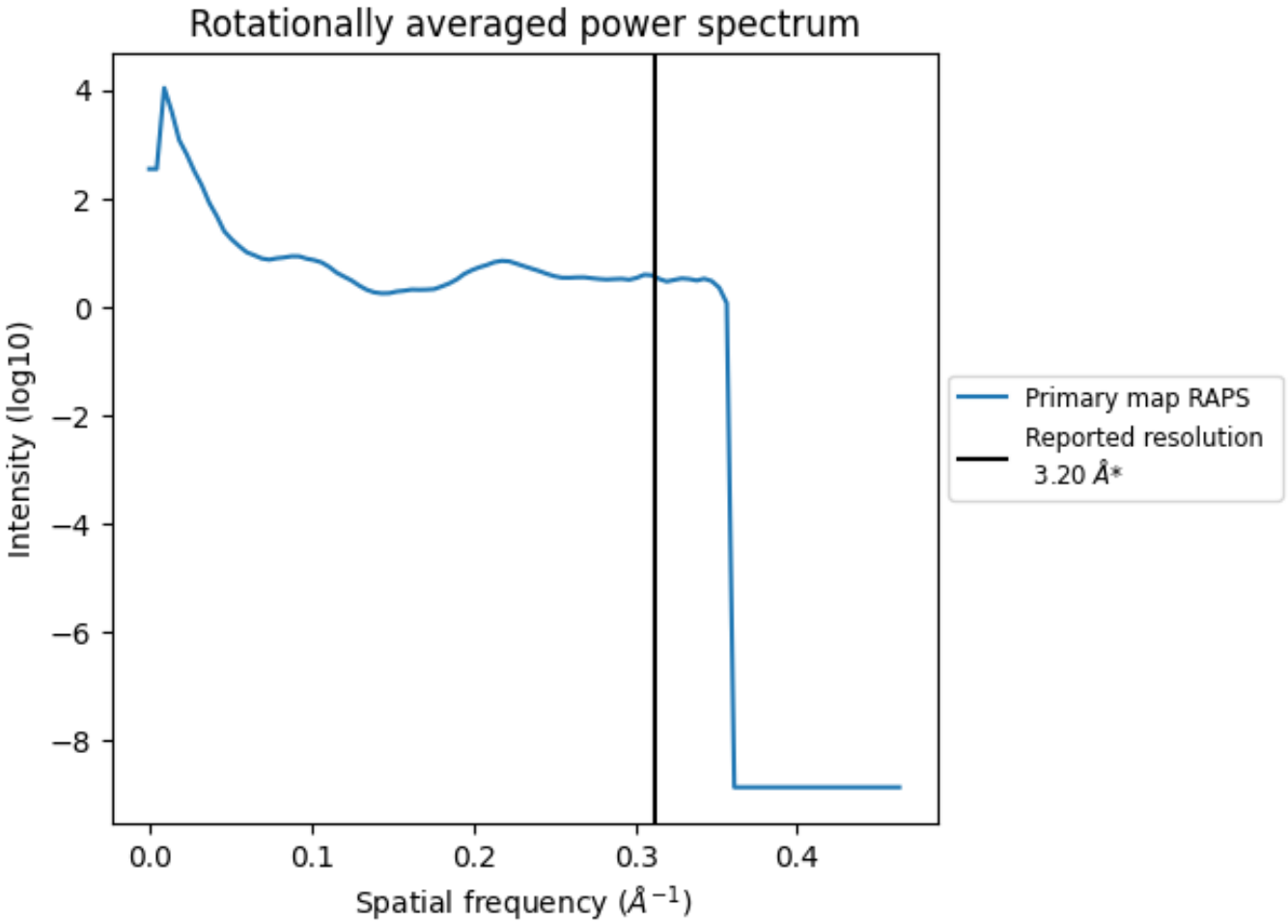
7.2 Volume estimate (i)



The volume at the recommended contour level is 39 nm^3; this corresponds to an approximate mass of 35 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)



*Reported resolution corresponds to spatial frequency of 0.312 Å^{-1}

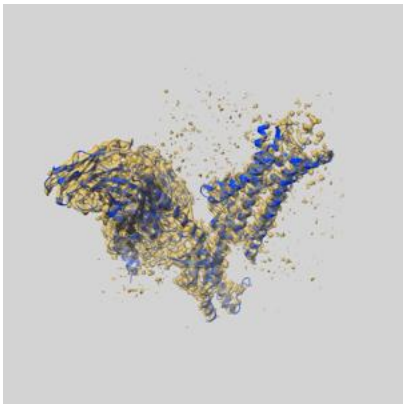
8 Fourier-Shell correlation (i)

This section was not generated. No FSC curve or half-maps provided.

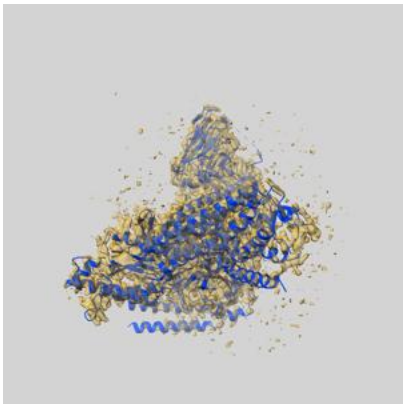
9 Map-model fit

This section contains information regarding the fit between EMDB map EMD-32313 and PDB model 7W53. Per-residue inclusion information can be found in section 3 on page 6.

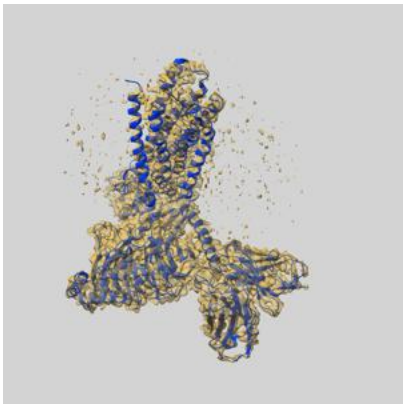
9.1 Map-model overlay



X



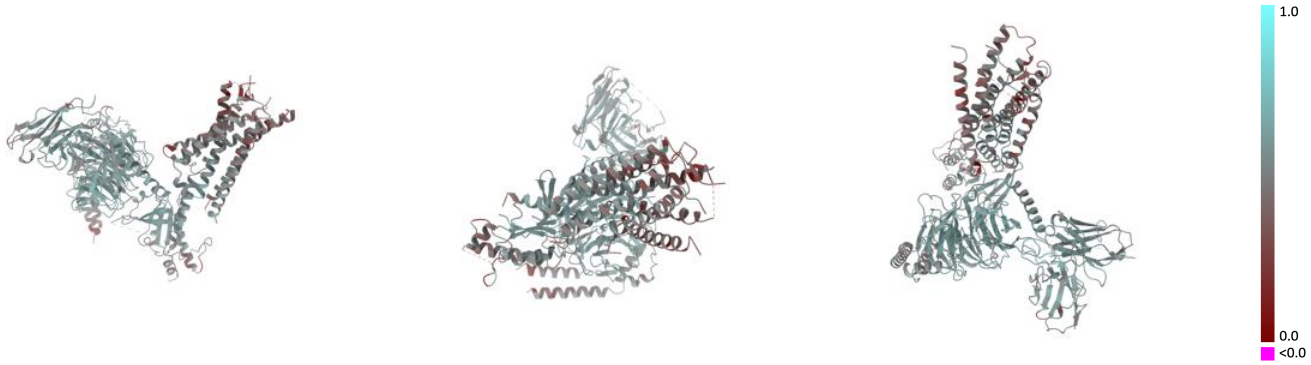
Y



Z

The images above show the 3D surface view of the map at the recommended contour level 0.024 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.

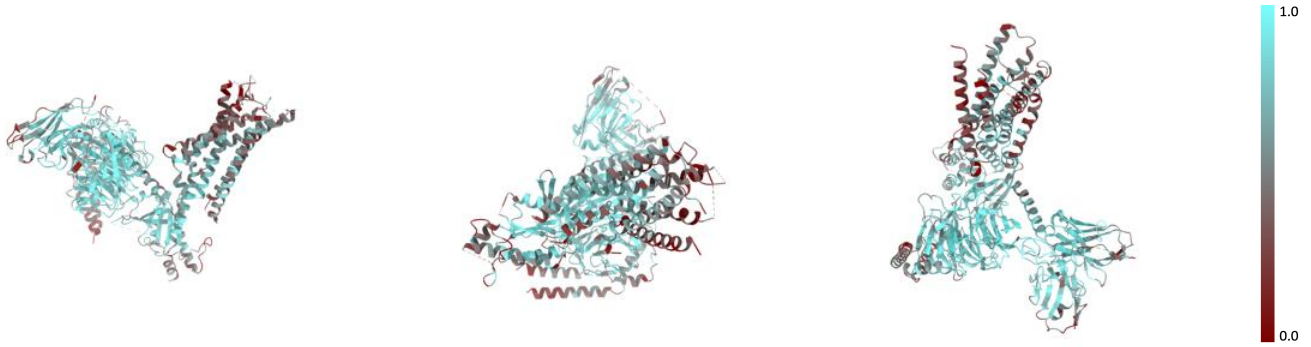
9.2 Q-score mapped to coordinate model (i)



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.

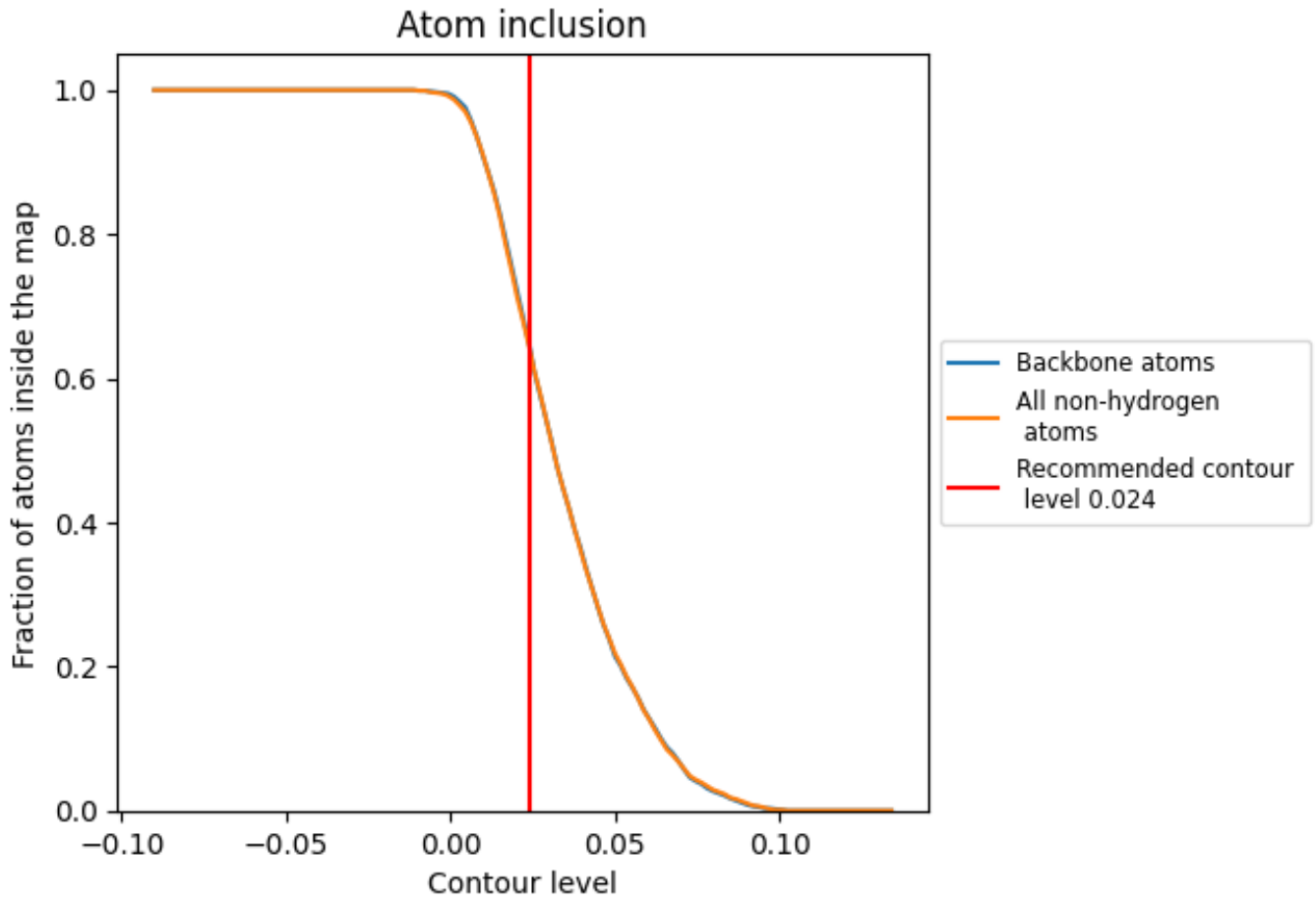
9.3 Atom inclusion mapped to coordinate model (i)



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 (0.024).

9.4 Atom inclusion (i)



At the recommended contour level, 64% of all backbone atoms, 64% of all non-hydrogen atoms, are inside the map.

9.5 Map-model fit summary

The table lists the average atom inclusion at the recommended contour level (0.024) and Q-score for the entire model and for each chain.

Chain	Atom inclusion	Q-score
All	0.6410	0.5140
A	0.6480	0.5090
B	0.7710	0.5680
C	0.5000	0.4700
E	0.7160	0.5410
G	0.5100	0.4790
R	0.4700	0.4360