wwPDB EM Validation Summary Report (i)

Feb 25, 2024 – 10:12 AM EST

PDB ID	:	5IPT
EMDB ID	:	EMD-8104
Title	:	Cryo-EM structure of GluN1/GluN2B NMDA receptor in the DCKA/D-APV-bound conformation, state 5
Authors	:	Zhu, S.; Stein, A.R.; Yoshioka, C.; Lee, C.H.; Goehring, A.; Mchaourab, S.H.; Gouaux, E.
Deposited on	:	2016-03-09
Resolution	:	14.10 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 (i) 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 (i)) were used in the production of this report:

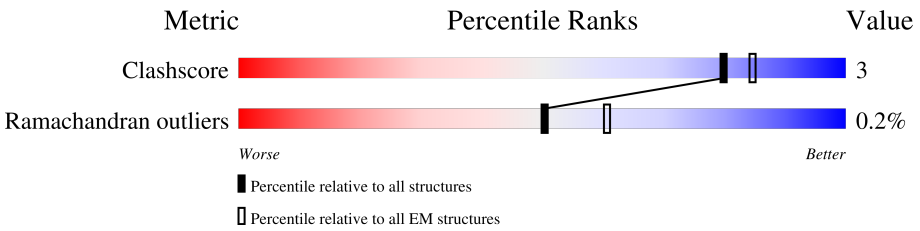
EMDB validation analysis	:	0.0.1.dev70
MolProbity	:	4.02b-467
Percentile statistics	:	20191225.v01 (using entries in the PDB archive December 25th 2019)
MapQ	:	1.9.13
Ideal geometry (proteins)	:	Engh & Huber (2001)
Ideal geometry (DNA, RNA)	:	Parkinson et al. (1996)
Validation Pipeline (wwPDB-VP)	:	2.36

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 14.10 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

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	822	16% 78% • 20%
1	C	822	7% 74% • 24%
2	B	825	18% 72% • 25%
2	D	825	12% 73% • 24%

2 Entry composition (i)

There are 2 unique types of molecules in this entry. The entry contains 10100 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 N-methyl-D-aspartate receptor subunit NR1-8a.

Mol	Chain	Residues	Atoms				AltConf	Trace
			Total	C	N	O		
1	A	657	2628	1314	657	657	0	0
1	C	626	2504	1252	626	626	0	0

There are 76 discrepancies between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	Comment	Reference
A	51	PHE	LYS	engineered mutation	UNP C0KD18
A	52	PHE	ARG	engineered mutation	UNP C0KD18
A	300	GLN	ASN	engineered mutation	UNP C0KD18
A	350	GLN	ASN	engineered mutation	UNP C0KD18
A	368	ASP	ASN	engineered mutation	UNP C0KD18
A	440	ASP	ASN	engineered mutation	UNP C0KD18
A	469	ASP	ASN	engineered mutation	UNP C0KD18
A	493	ALA	LYS	engineered mutation	UNP C0KD18
A	494	ALA	LYS	engineered mutation	UNP C0KD18
A	495	ALA	GLU	engineered mutation	UNP C0KD18
A	?	-	LYS	deletion	UNP C0KD18
A	?	-	VAL	deletion	UNP C0KD18
A	?	-	ASN	deletion	UNP C0KD18
A	?	-	SER	deletion	UNP C0KD18
A	?	-	GLU	deletion	UNP C0KD18
A	?	-	GLU	deletion	UNP C0KD18
A	?	-	GLU	deletion	UNP C0KD18
A	?	-	GLU	deletion	UNP C0KD18
A	602	ARG	GLY	engineered mutation	UNP C0KD18
A	609	LEU	ILE	engineered mutation	UNP C0KD18
A	648	ARG	ASP	engineered mutation	UNP C0KD18
A	761	GLU	ASN	engineered mutation	UNP C0KD18
A	829	SER	-	expression tag	UNP C0KD18
A	830	ARG	-	expression tag	UNP C0KD18
A	831	ALA	-	expression tag	UNP C0KD18
A	832	GLU	-	expression tag	UNP C0KD18

Continued on next page...

Continued from previous page...

Chain	Residue	Modelled	Actual	Comment	Reference
A	833	ALA	-	expression tag	UNP C0KD18
A	834	LYS	-	expression tag	UNP C0KD18
A	835	ARG	-	expression tag	UNP C0KD18
A	836	MET	-	expression tag	UNP C0KD18
A	837	LYS	-	expression tag	UNP C0KD18
A	838	GLY	-	expression tag	UNP C0KD18
A	839	LEU	-	expression tag	UNP C0KD18
A	840	GLU	-	expression tag	UNP C0KD18
A	841	VAL	-	expression tag	UNP C0KD18
A	842	LEU	-	expression tag	UNP C0KD18
A	843	PHE	-	expression tag	UNP C0KD18
A	844	GLN	-	expression tag	UNP C0KD18
C	51	PHE	LYS	engineered mutation	UNP C0KD18
C	52	PHE	ARG	engineered mutation	UNP C0KD18
C	300	GLN	ASN	engineered mutation	UNP C0KD18
C	350	GLN	ASN	engineered mutation	UNP C0KD18
C	368	ASP	ASN	engineered mutation	UNP C0KD18
C	440	ASP	ASN	engineered mutation	UNP C0KD18
C	469	ASP	ASN	engineered mutation	UNP C0KD18
C	493	ALA	LYS	engineered mutation	UNP C0KD18
C	494	ALA	LYS	engineered mutation	UNP C0KD18
C	495	ALA	GLU	engineered mutation	UNP C0KD18
C	?	-	LYS	deletion	UNP C0KD18
C	?	-	VAL	deletion	UNP C0KD18
C	?	-	ASN	deletion	UNP C0KD18
C	?	-	SER	deletion	UNP C0KD18
C	?	-	GLU	deletion	UNP C0KD18
C	?	-	GLU	deletion	UNP C0KD18
C	?	-	GLU	deletion	UNP C0KD18
C	?	-	GLU	deletion	UNP C0KD18
C	602	ARG	GLY	engineered mutation	UNP C0KD18
C	609	LEU	ILE	engineered mutation	UNP C0KD18
C	648	ARG	ASP	engineered mutation	UNP C0KD18
C	761	GLU	ASN	engineered mutation	UNP C0KD18
C	829	SER	-	expression tag	UNP C0KD18
C	830	ARG	-	expression tag	UNP C0KD18
C	831	ALA	-	expression tag	UNP C0KD18
C	832	GLU	-	expression tag	UNP C0KD18
C	833	ALA	-	expression tag	UNP C0KD18
C	834	LYS	-	expression tag	UNP C0KD18
C	835	ARG	-	expression tag	UNP C0KD18
C	836	MET	-	expression tag	UNP C0KD18

Continued on next page...

Continued from previous page...

Chain	Residue	Modelled	Actual	Comment	Reference
C	837	LYS	-	expression tag	UNP C0KD18
C	838	GLY	-	expression tag	UNP C0KD18
C	839	LEU	-	expression tag	UNP C0KD18
C	840	GLU	-	expression tag	UNP C0KD18
C	841	VAL	-	expression tag	UNP C0KD18
C	842	LEU	-	expression tag	UNP C0KD18
C	843	PHE	-	expression tag	UNP C0KD18
C	844	GLN	-	expression tag	UNP C0KD18

- Molecule 2 is a protein called Ionotropic glutamate receptor subunit NR2B.

Mol	Chain	Residues	Atoms				AltConf	Trace
			Total	C	N	O		
2	B	615	2460	1230	615	615	0	0
			Total	C	N	O		
2	D	627	2508	1254	627	627	0	0

There are 48 discrepancies between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	Comment	Reference
B	20	SER	MET	engineered mutation	UNP A7XY94
B	21	ARG	GLY	engineered mutation	UNP A7XY94
B	22	ALA	CYS	engineered mutation	UNP A7XY94
B	64	GLU	ALA	engineered mutation	UNP A7XY94
B	69	GLN	ASN	engineered mutation	UNP A7XY94
B	343	ASP	ASN	engineered mutation	UNP A7XY94
B	?	-	LYS	deletion	UNP A7XY94
B	?	-	TYR	deletion	UNP A7XY94
B	?	-	TYR	deletion	UNP A7XY94
B	?	-	VAL	deletion	UNP A7XY94
B	486	VAL	THR	engineered mutation	UNP A7XY94
B	?	-	ARG	deletion	UNP A7XY94
B	?	-	CYS	deletion	UNP A7XY94
B	?	-	LEU	deletion	UNP A7XY94
B	?	-	ALA	deletion	UNP A7XY94
B	?	-	ASP	deletion	UNP A7XY94
B	?	-	GLY	deletion	UNP A7XY94
B	?	-	ARG	deletion	UNP A7XY94
B	?	-	GLU	deletion	UNP A7XY94
B	?	-	PRO	deletion	UNP A7XY94
B	?	-	GLY	deletion	UNP A7XY94
B	601	LEU	VAL	engineered mutation	UNP A7XY94

Continued on next page...

Continued from previous page...

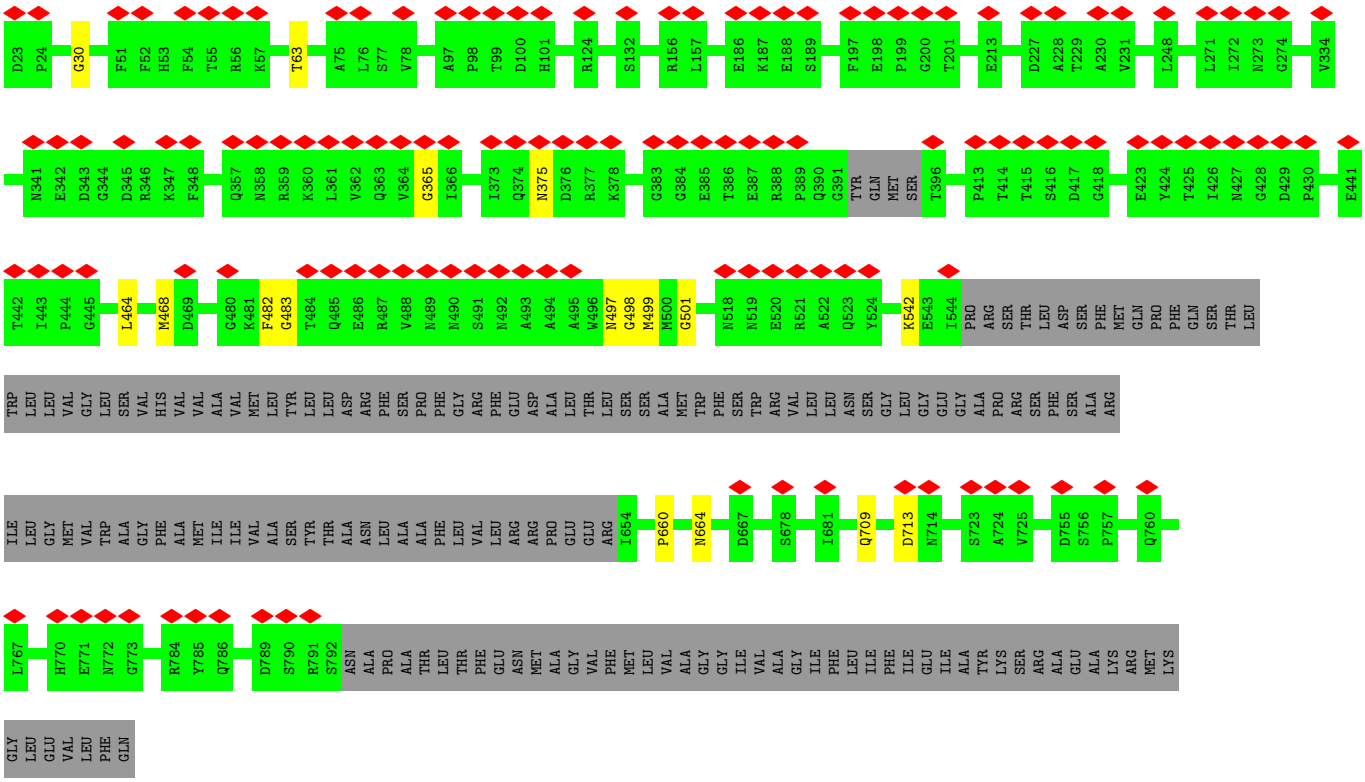
Chain	Residue	Modelled	Actual	Comment	Reference
B	640	ARG	GLU	engineered mutation	UNP A7XY94
B	641	ARG	GLU	engineered mutation	UNP A7XY94
D	20	SER	MET	engineered mutation	UNP A7XY94
D	21	ARG	GLY	engineered mutation	UNP A7XY94
D	22	ALA	CYS	engineered mutation	UNP A7XY94
D	64	GLU	ALA	engineered mutation	UNP A7XY94
D	69	GLN	ASN	engineered mutation	UNP A7XY94
D	343	ASP	ASN	engineered mutation	UNP A7XY94
D	?	-	LYS	deletion	UNP A7XY94
D	?	-	TYR	deletion	UNP A7XY94
D	?	-	TYR	deletion	UNP A7XY94
D	?	-	VAL	deletion	UNP A7XY94
D	486	VAL	THR	engineered mutation	UNP A7XY94
D	?	-	ARG	deletion	UNP A7XY94
D	?	-	CYS	deletion	UNP A7XY94
D	?	-	LEU	deletion	UNP A7XY94
D	?	-	ALA	deletion	UNP A7XY94
D	?	-	ASP	deletion	UNP A7XY94
D	?	-	GLY	deletion	UNP A7XY94
D	?	-	ARG	deletion	UNP A7XY94
D	?	-	GLU	deletion	UNP A7XY94
D	?	-	PRO	deletion	UNP A7XY94
D	?	-	GLY	deletion	UNP A7XY94
D	601	LEU	VAL	engineered mutation	UNP A7XY94
D	640	ARG	GLU	engineered mutation	UNP A7XY94
D	641	ARG	GLU	engineered mutation	UNP A7XY94

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.

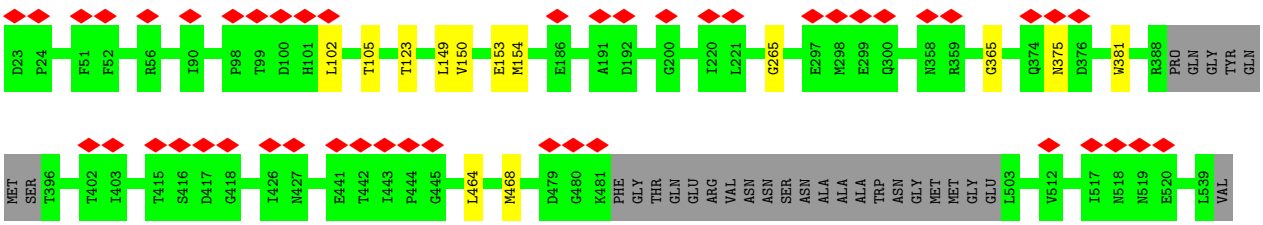
- Molecule 1: N-methyl-D-aspartate receptor subunit NR1-8a

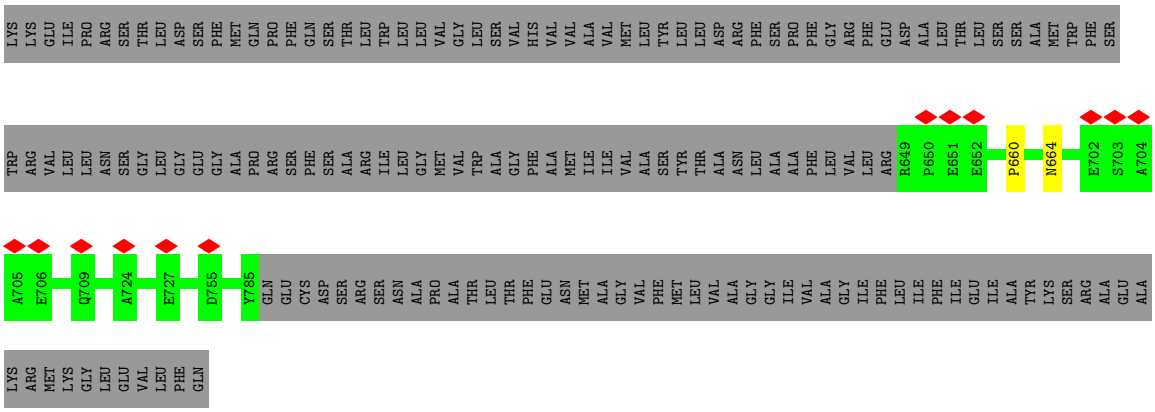
Chain A:



- Molecule 1: N-methyl-D-aspartate receptor subunit NR1-8a

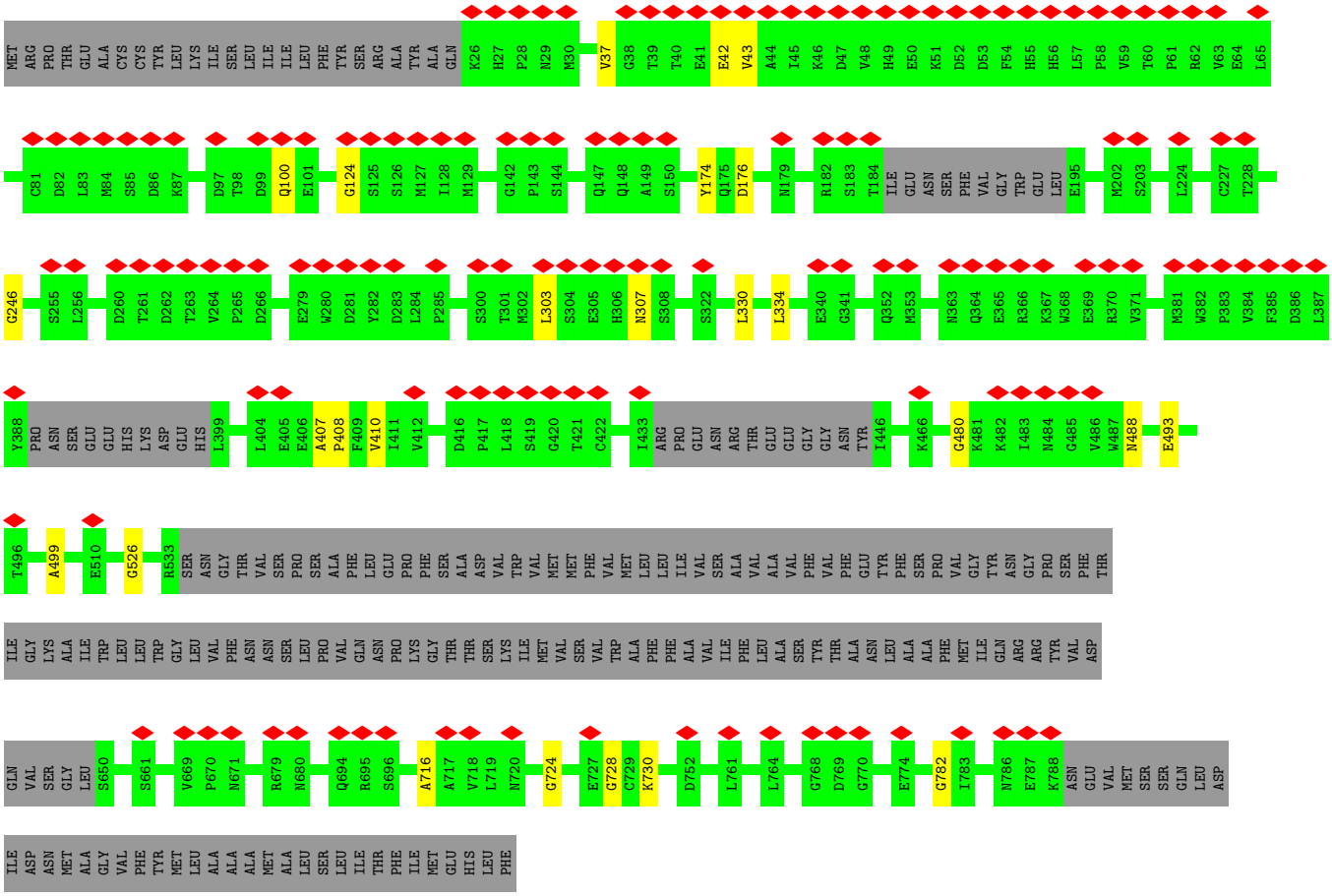
Chain C:





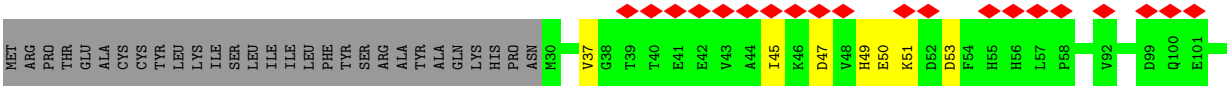
• Molecule 2: Ionotropic glutamate receptor subunit NR2B

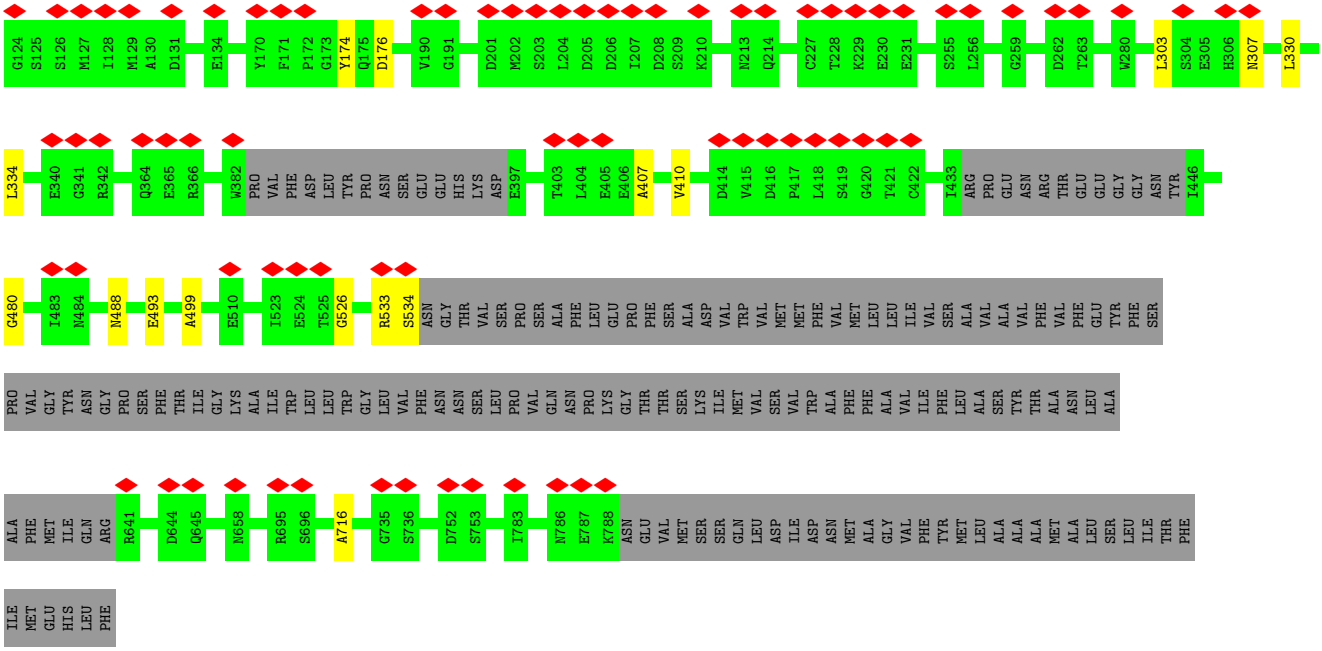
Chain B:



• Molecule 2: Ionotropic glutamate receptor subunit NR2B

Chain D:




G124 S125 S126 M127 I128 M129 A130 D131 E134 Y170 F171 P172 G173 Y174 Q175 D176 V190 G191 D201 M202 S203 L204 D205 D206 I207 D208 S209 K210 N213 Q214 C227 T228 K229 E230 E231 S255 L256 G259 D262 T263 W280 L303 S304 E305 H306 N307 L330
L334 E340 G341 R342 Q364 E365 R366 W382 PRO VAL PHE ASP LEU TYR PRO ASN SER GLU GLU HIS LYS ASP E397 T403 L404 E405 E406 A407 V410 D414 V415 D416 P417 L418 S419 G420 T421 C422 I433 ARG PRO GLU ASN ARG THR GLU GLU GLY GLY ASN TYR I446
G480 I483 M484 N488 E493 A499 E510 I523 E524 T525 G526 R533 S534 ASN GLY THR VAL SER PRO SER ALA PHE LEU GLU PRO PHE SER ALA ASP VAL TRP VAL MET MET PHE VAL MET LEU LEU ILE VAL SER ALA VAL ALA VAL PHE VAL PHE GLU TYR PHE SER
PRO VAL GLY TYR ASN GLY PRO SER PHE THR ILE GLY LYS ALA ILE TRP LEU LEU TRP GLY LEU VAL PHE ASN ASN SER LEU PRO VAL GLN ASN PRO LYS GLY THR THR SER LYS ILE MET VAL SER VAL TRP ALA PHE PHE ALA VAL ILE PHE LEU ALA SER TYR THR ALA ASN LEU ALA
ALA PHE MET ILE GLN ARG R641 D644 Q645 N658 R695 S696 A716 G735 S736 D752 S753 I783 N786 E787 K788 ASN GLU VAL MET SER SER GLN LEU ASP ILE ASP ASN MET ALA GLY VAL PHE TYR MET LEU ALA ALA ALA MET ALA LEU SER LEU ILE THR PHE
ILE MET GLU HIS LEU PHE

4 Experimental information (i)

Property	Value	Source
EM reconstruction method	SINGLE PARTICLE	Depositor
Imposed symmetry	POINT, C1	Depositor
Number of particles used	32608	Depositor
Resolution determination method	FSC 0.143 CUT-OFF	Depositor
CTF correction method	PHASE FLIPPING AND AMPLITUDE CORRECTION	Depositor
Microscope	FEI TITAN KRIOS	Depositor
Voltage (kV)	300	Depositor
Electron dose ($e^-/Å^2$)	10.5	Depositor
Minimum defocus (nm)	1500	Depositor
Maximum defocus (nm)	3000	Depositor
Magnification	Not provided	
Image detector	GATAN K2 SUMMIT (4k x 4k)	Depositor
Maximum map value	0.152	Depositor
Minimum map value	-0.052	Depositor
Average map value	0.006	Depositor
Map value standard deviation	0.020	Depositor
Recommended contour level	0.052	Depositor
Map size (Å)	236.47003, 215.07004, 204.37003	wwPDB
Map dimensions	221, 201, 191	wwPDB
Map angles (°)	90.0, 90.0, 90.0	wwPDB
Pixel spacing (Å)	1.0700002, 1.0700002, 1.0700002	Depositor

5 Model quality (i)

5.1 Standard geometry (i)

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	0.29	0/2625	0.63	0/3276
1	C	0.29	0/2500	0.64	1/3118 (0.0%)
2	B	0.30	0/2455	0.66	2/3060 (0.1%)
2	D	0.32	0/2504	0.68	1/3123 (0.0%)
All	All	0.30	0/10084	0.65	4/12577 (0.0%)

Chiral center outliers are detected by calculating the chiral volume of a chiral center and verifying if the center is modelled as a planar moiety or with the opposite hand.A planarity outlier is detected by checking planarity of atoms in a peptide group, atoms in a mainchain group or atoms of a sidechain that are expected to be planar.

Mol	Chain	#Chirality outliers	#Planarity outliers
1	A	0	1
2	B	0	1
All	All	0	2

There are no bond length outliers.

All (4) bond angle outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(^{o})	Ideal(^{o})
2	B	37	VAL	C-N-CA	-6.65	108.34	122.30
1	C	123	THR	N-CA-C	5.61	126.13	111.00
2	B	42	GLU	N-CA-C	-5.21	96.92	111.00
2	D	37	VAL	C-N-CA	5.12	133.05	122.30

There are no chirality outliers.

All (2) planarity outliers are listed below:

Mol	Chain	Res	Type	Group
1	A	542	LYS	Peptide
2	B	246	GLY	Peptide

5.2 Too-close contacts (i)

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	2628	0	704	8	0
1	C	2504	0	664	8	0
2	B	2460	0	666	9	0
2	D	2508	0	685	10	0
All	All	10100	0	2719	35	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 3.

The worst 5 of 35 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)
1:C:660:PRO:O	1:C:664:ASN:N	2.15	0.79
2:D:50:GLU:O	2:D:53:ASP:N	2.20	0.75
1:C:464:LEU:O	1:C:468:MET:N	2.20	0.74
1:A:660:PRO:O	1:A:664:ASN:N	2.22	0.73
1:A:483:GLY:HA2	1:A:497:ASN:O	1.93	0.69

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	651/822 (79%)	630 (97%)	21 (3%)	0	100	100
1	C	618/822 (75%)	597 (97%)	21 (3%)	0	100	100

Continued on next page...

Continued from previous page...

Mol	Chain	Analysed	Favoured	Allowed	Outliers	Percentiles	
2	B	605/825 (73%)	574 (95%)	27 (4%)	4 (1%)	22	63
2	D	619/825 (75%)	592 (96%)	25 (4%)	2 (0%)	41	77
All	All	2493/3294 (76%)	2393 (96%)	94 (4%)	6 (0%)	50	81

5 of 6 Ramachandran outliers are listed below:

Mol	Chain	Res	Type
2	B	407	ALA
2	B	408	PRO
2	D	407	ALA
2	D	410	VAL
2	B	410	VAL

5.3.2 Protein sidechains (i)

There are no protein residues with a non-rotameric sidechain to report in this entry.

5.3.3 RNA (i)

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 monosaccharides 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 (i)

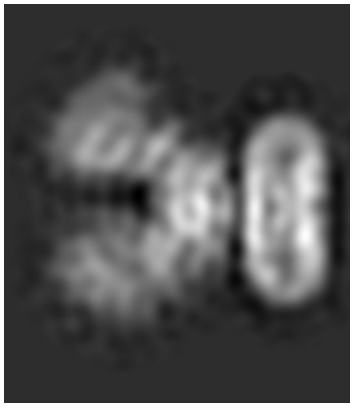
This section contains visualisations of the EMDB entry EMD-8104. These allow visual inspection of the internal detail of the map and identification of artifacts.

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.

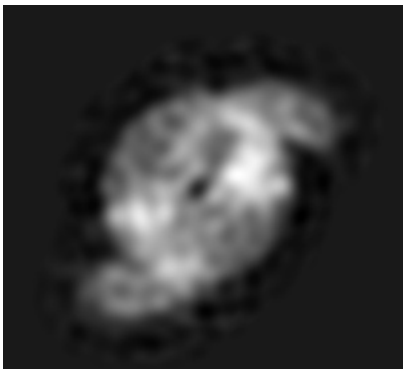
6.1 Orthogonal projections (i)

6.1.1 Primary map

X



Y

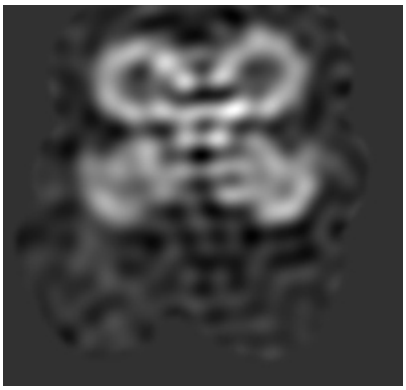


Z

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

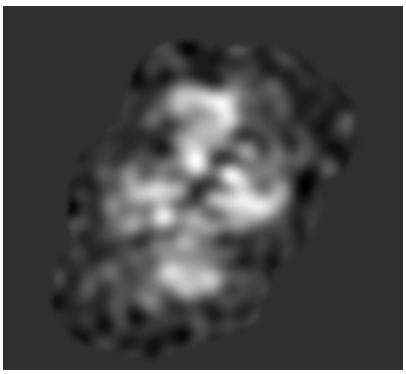
6.2 Central slices (i)

6.2.1 Primary map



X Index: 110

Y Index: 100



Z Index: 95

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: 137

Y Index: 99

Z Index: 138

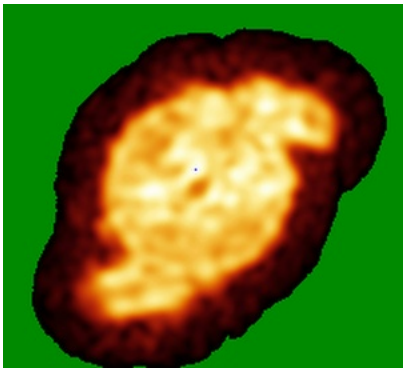
The images above show the largest variance slices of the map in three orthogonal directions.

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

6.4.1 Primary map

X

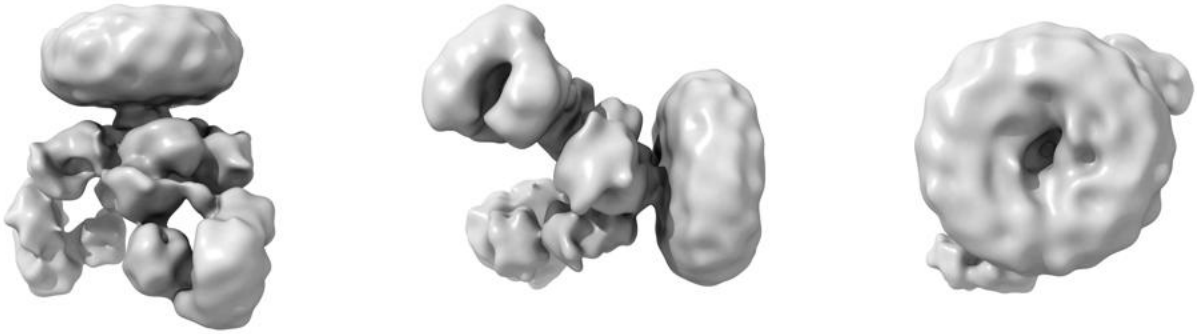
Y


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 0.052. These images, in conjunction with the slice images, may facilitate assessment of whether an appropriate contour level has been provided.

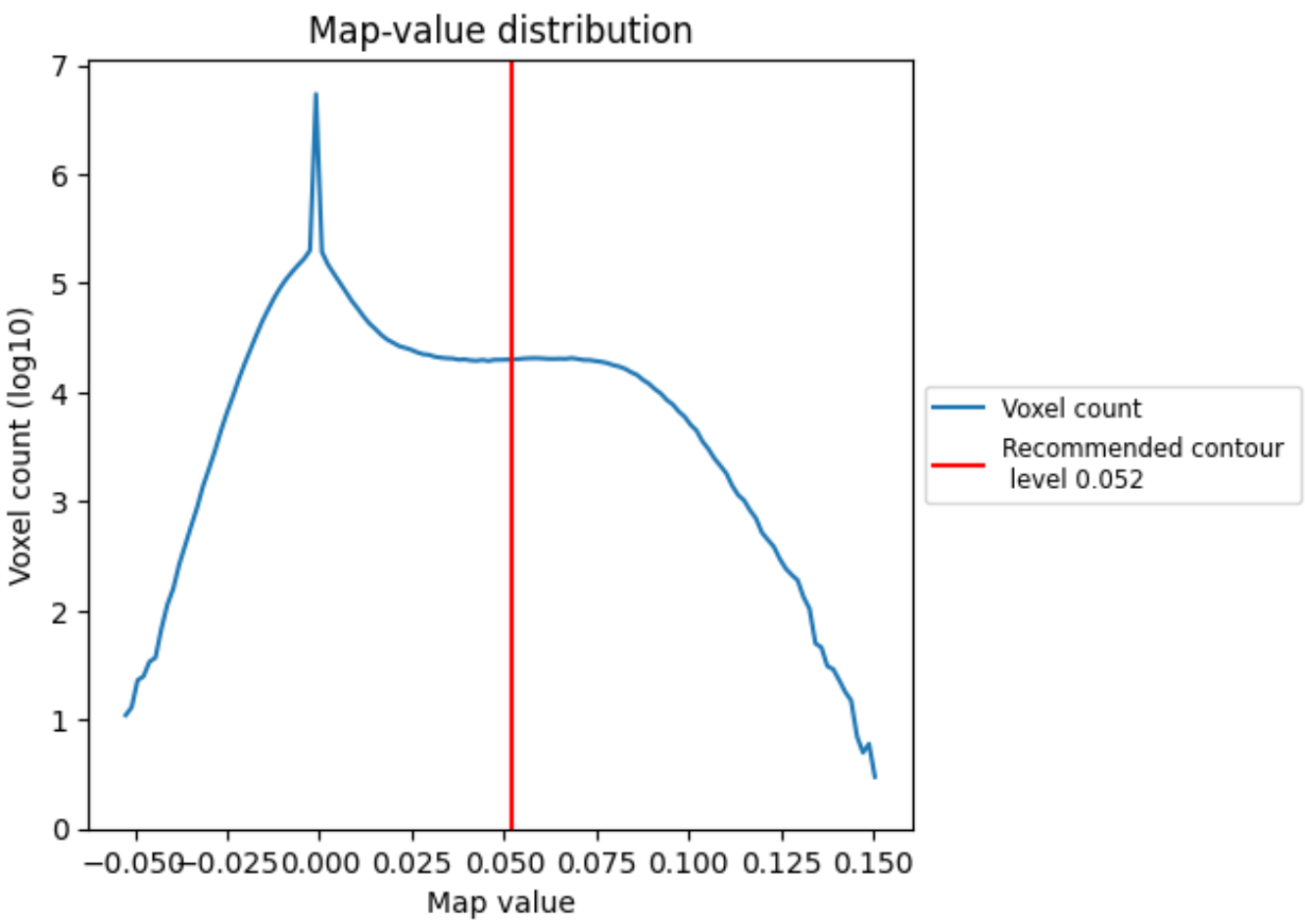
6.6 Mask visualisation (i)

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

7 Map analysis (i)

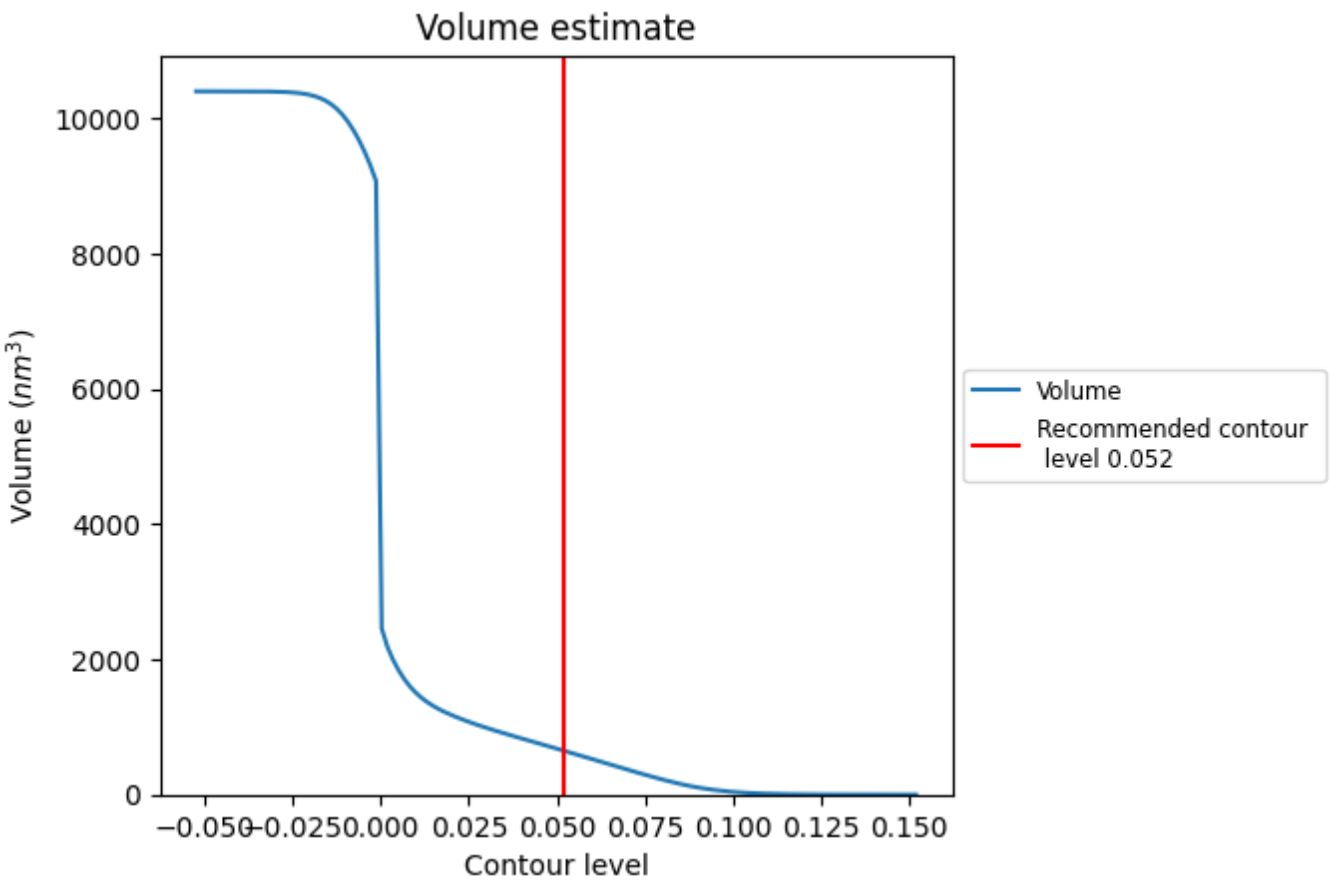
This section contains the results of statistical analysis of the map.

7.1 Map-value distribution (i)



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 648 nm^3; this corresponds to an approximate mass of 586 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.

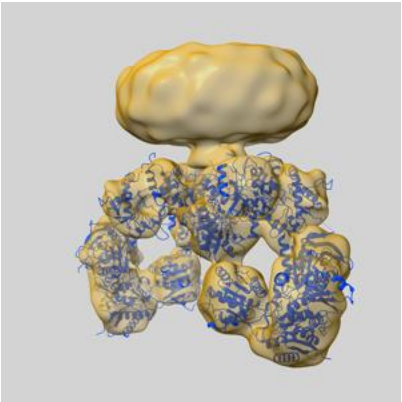
8 Fourier-Shell correlation (i)

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

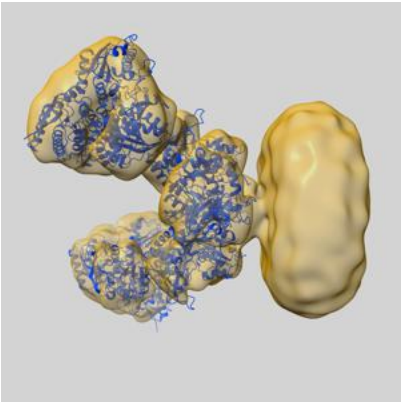
9 Map-model fit (i)

This section contains information regarding the fit between EMDB map EMD-8104 and PDB model 5IPT. Per-residue inclusion information can be found in section 3 on page 7.

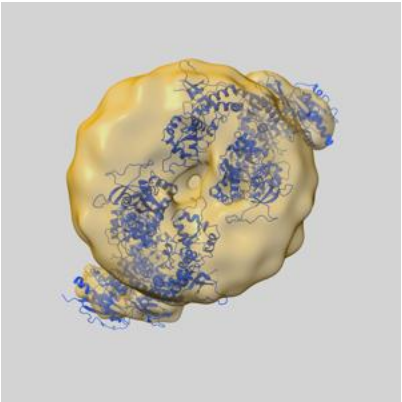
9.1 Map-model overlay (i)



X



Y



Z

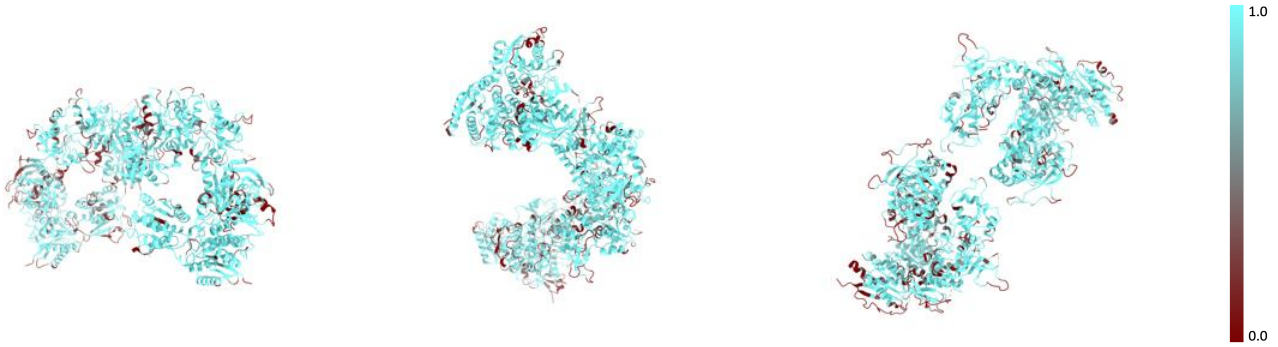
The images above show the 3D surface view of the map at the recommended contour level 0.052 at 50% transparency in yellow overlaid with a ribbon representation of the model coloured in blue. These images allow for the visual assessment of the quality of fit between the atomic model and the map.

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.

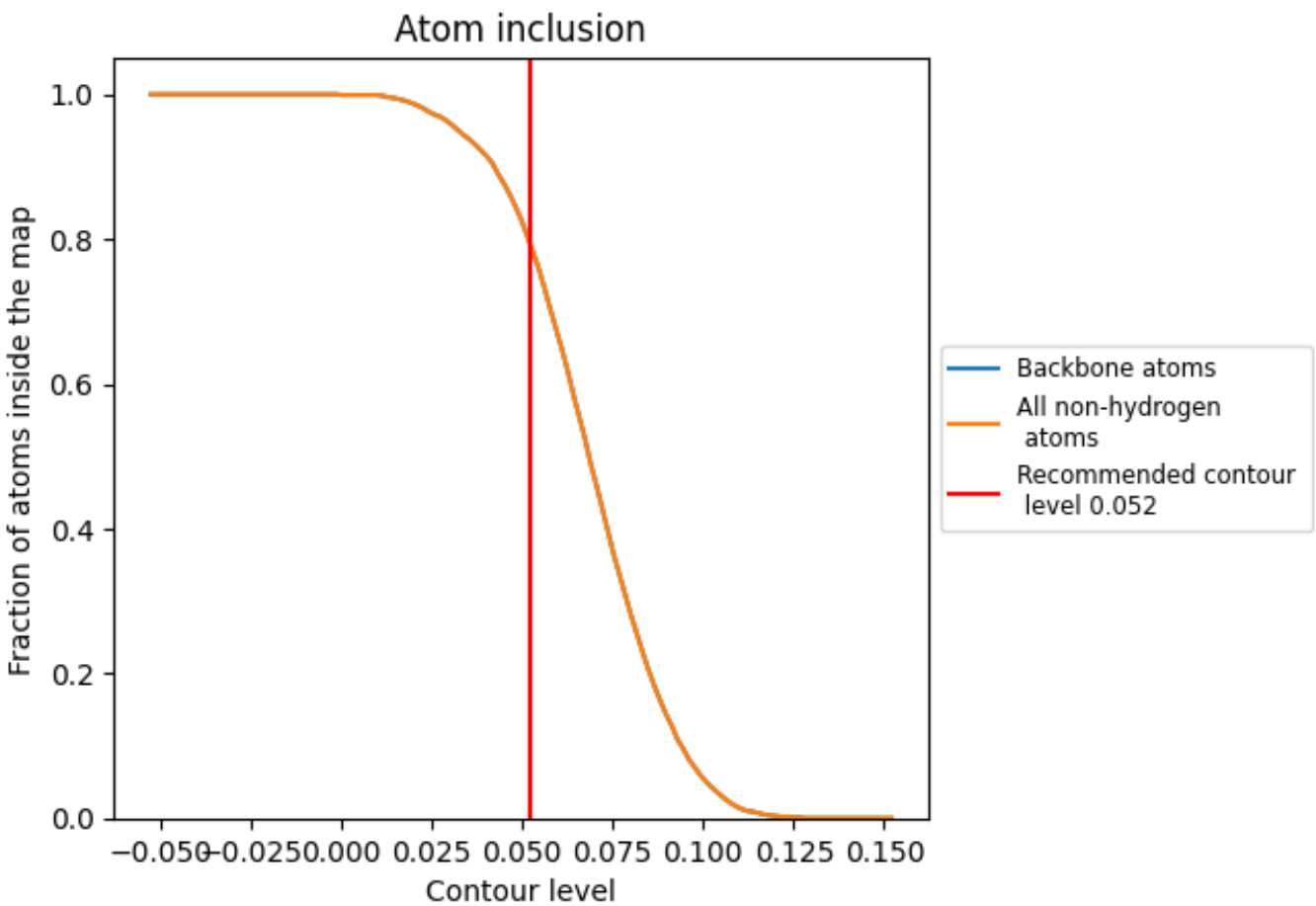
9.3 Atom inclusion mapped to coordinate model (i)



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 (0.052).

9.4 Atom inclusion (i)



At the recommended contour level, 80% of all backbone atoms, 80% 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 (0.052) and Q-score for the entire model and for each chain.

Chain	Atom inclusion	Q-score
All	0.7970	0.0800
A	0.7680	0.0730
B	0.7210	0.0700
C	0.8830	0.0980
D	0.8170	0.0810