wwPDB EM Validation Summary Report (i)

Mar 4, 2024 – 10:38 AM EST

PDB ID	:	6DBI
EMDB ID	:	EMD-7843
Title	:	Cryo-EM structure of RAG in complex with 12-RSS and 23-RSS nicked DNA intermediates
Authors	:	Wu, H.; Liao, M.; Ru, H.; Mi, W.
Deposited on	:	2018-05-03
Resolution	:	3.40 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:

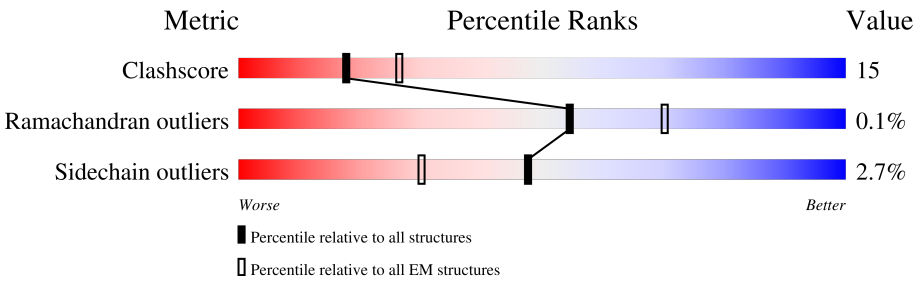
EMDB validation analysis	:	0.0.1.dev70
MolProbity	:	4.02b-467
Percentile statistics	:	20191225.v01 (using entries in the PDB archive December 25th 2019)
MapQ	:	1.9.13
Ideal geometry (proteins)	:	Engh & Huber (2001)
Ideal geometry (DNA, RNA)	:	Parkinson et al. (1996)
Validation Pipeline (wwPDB-VP)	:	2.36

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 3.40 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	1159	8% 38% 15% • 46%
1	C	1159	8% 39% 14% • 46%
2	B	533	• 40% 25% • 34%
2	D	533	5% 45% 20% • 34%
3	E	34	50% 29% 68% •
4	F	50	44% 40% 60%
5	G	61	54% 46% 54%
6	H	45	67% 58% 42%

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Mol	Chain	Length	Quality of chain
7	I	16	12% 44% 56%
7	J	16	19% 50% 50%

2 Entry composition (i)

There are 9 unique types of molecules in this entry. The entry contains 20001 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 Recombination activating gene 1 - MBP chimera.

Mol	Chain	Residues	Atoms					AltConf	Trace
			Total	C	N	O	S		
1	C	622	5008	3134	901	936	37	0	0
1	A	622	5008	3134	901	936	37	0	0

There are 68 discrepancies between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	Comment	Reference
C	-127	MET	-	initiating methionine	UNP P0AEX9
C	-126	GLY	-	expression tag	UNP P0AEX9
C	-125	SER	-	expression tag	UNP P0AEX9
C	-124	SER	-	expression tag	UNP P0AEX9
C	-123	HIS	-	expression tag	UNP P0AEX9
C	-122	HIS	-	expression tag	UNP P0AEX9
C	-121	HIS	-	expression tag	UNP P0AEX9
C	-120	HIS	-	expression tag	UNP P0AEX9
C	-119	HIS	-	expression tag	UNP P0AEX9
C	-118	HIS	-	expression tag	UNP P0AEX9
C	-117	GLY	-	expression tag	UNP P0AEX9
C	-116	THR	-	expression tag	UNP P0AEX9
C	-115	LYS	-	expression tag	UNP P0AEX9
C	-114	THR	-	expression tag	UNP P0AEX9
C	251	GLY	-	linker	UNP P0AEX9
C	252	THR	-	linker	UNP P0AEX9
C	253	ASP	-	linker	UNP P0AEX9
C	254	TYR	-	linker	UNP P0AEX9
C	255	ASP	-	linker	UNP P0AEX9
C	256	ILE	-	linker	UNP P0AEX9
C	257	PRO	-	linker	UNP P0AEX9
C	258	THR	-	linker	UNP P0AEX9
C	259	THR	-	linker	UNP P0AEX9
C	260	LEU	-	linker	UNP P0AEX9
C	261	GLU	-	linker	UNP P0AEX9
C	262	VAL	-	linker	UNP P0AEX9

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Chain	Residue	Modelled	Actual	Comment	Reference
C	263	LEU	-	linker	UNP P0AEX9
C	264	PHE	-	linker	UNP P0AEX9
C	265	GLN	-	linker	UNP P0AEX9
C	266	GLY	-	linker	UNP P0AEX9
C	267	PRO	-	linker	UNP P0AEX9
C	268	LEU	-	linker	UNP P0AEX9
C	269	GLY	-	linker	UNP P0AEX9
C	270	SER	-	linker	UNP P0AEX9
A	-127	MET	-	initiating methionine	UNP P0AEX9
A	-126	GLY	-	expression tag	UNP P0AEX9
A	-125	SER	-	expression tag	UNP P0AEX9
A	-124	SER	-	expression tag	UNP P0AEX9
A	-123	HIS	-	expression tag	UNP P0AEX9
A	-122	HIS	-	expression tag	UNP P0AEX9
A	-121	HIS	-	expression tag	UNP P0AEX9
A	-120	HIS	-	expression tag	UNP P0AEX9
A	-119	HIS	-	expression tag	UNP P0AEX9
A	-118	HIS	-	expression tag	UNP P0AEX9
A	-117	GLY	-	expression tag	UNP P0AEX9
A	-116	THR	-	expression tag	UNP P0AEX9
A	-115	LYS	-	expression tag	UNP P0AEX9
A	-114	THR	-	expression tag	UNP P0AEX9
A	251	GLY	-	linker	UNP P0AEX9
A	252	THR	-	linker	UNP P0AEX9
A	253	ASP	-	linker	UNP P0AEX9
A	254	TYR	-	linker	UNP P0AEX9
A	255	ASP	-	linker	UNP P0AEX9
A	256	ILE	-	linker	UNP P0AEX9
A	257	PRO	-	linker	UNP P0AEX9
A	258	THR	-	linker	UNP P0AEX9
A	259	THR	-	linker	UNP P0AEX9
A	260	LEU	-	linker	UNP P0AEX9
A	261	GLU	-	linker	UNP P0AEX9
A	262	VAL	-	linker	UNP P0AEX9
A	263	LEU	-	linker	UNP P0AEX9
A	264	PHE	-	linker	UNP P0AEX9
A	265	GLN	-	linker	UNP P0AEX9
A	266	GLY	-	linker	UNP P0AEX9
A	267	PRO	-	linker	UNP P0AEX9
A	268	LEU	-	linker	UNP P0AEX9
A	269	GLY	-	linker	UNP P0AEX9
A	270	SER	-	linker	UNP P0AEX9

- Molecule 2 is a protein called Recombination activating gene 2.

Mol	Chain	Residues	Atoms					AltConf	Trace
			Total	C	N	O	S		
2	D	351	2714	1716	470	509	19	0	0
2	B	351	2714	1716	470	509	19	0	0

There are 6 discrepancies between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	Comment	Reference
D	-2	GLY	-	expression tag	UNP Q1RLW7
D	-1	GLY	-	expression tag	UNP Q1RLW7
D	0	SER	-	expression tag	UNP Q1RLW7
B	-2	GLY	-	expression tag	UNP Q1RLW7
B	-1	GLY	-	expression tag	UNP Q1RLW7
B	0	SER	-	expression tag	UNP Q1RLW7

- Molecule 3 is a DNA chain called Forward strand of 12-RSS signal end.

Mol	Chain	Residues	Atoms					AltConf	Trace
			Total	C	N	O	P		
3	E	34	697	330	138	195	34	0	0

- Molecule 4 is a DNA chain called Reverse strand of 12-RSS.

Mol	Chain	Residues	Atoms					AltConf	Trace
			Total	C	N	O	P		
4	F	50	1027	489	183	305	50	0	0

- Molecule 5 is a DNA chain called Reverse strand of 23-RSS.

Mol	Chain	Residues	Atoms					AltConf	Trace
			Total	C	N	O	P		
5	G	61	1256	596	235	364	61	0	0

- Molecule 6 is a DNA chain called Forward strand of 23-RSS signal end.

Mol	Chain	Residues	Atoms					AltConf	Trace
			Total	C	N	O	P		
6	H	45	919	437	169	268	45	0	0

- Molecule 7 is a DNA chain called Forward strand of coding flank.

Mol	Chain	Residues	Atoms					AltConf	Trace
			Total	C	N	O	P		
7	I	16	326	156	54	100	16	0	0
7	J	16	326	156	54	100	16	0	0

- Molecule 8 is ZINC ION (three-letter code: ZN) (formula: Zn).

Mol	Chain	Residues	Atoms		AltConf
			Total	Zn	
8	C	1	1	1	0
8	A	1	1	1	0

- Molecule 9 is CALCIUM ION (three-letter code: CA) (formula: Ca).

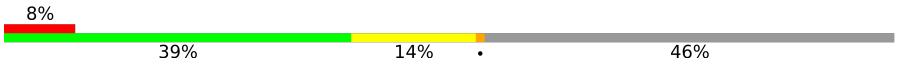
Mol	Chain	Residues	Atoms		AltConf
			Total	Ca	
9	C	2	2	2	0
9	A	2	2	2	0

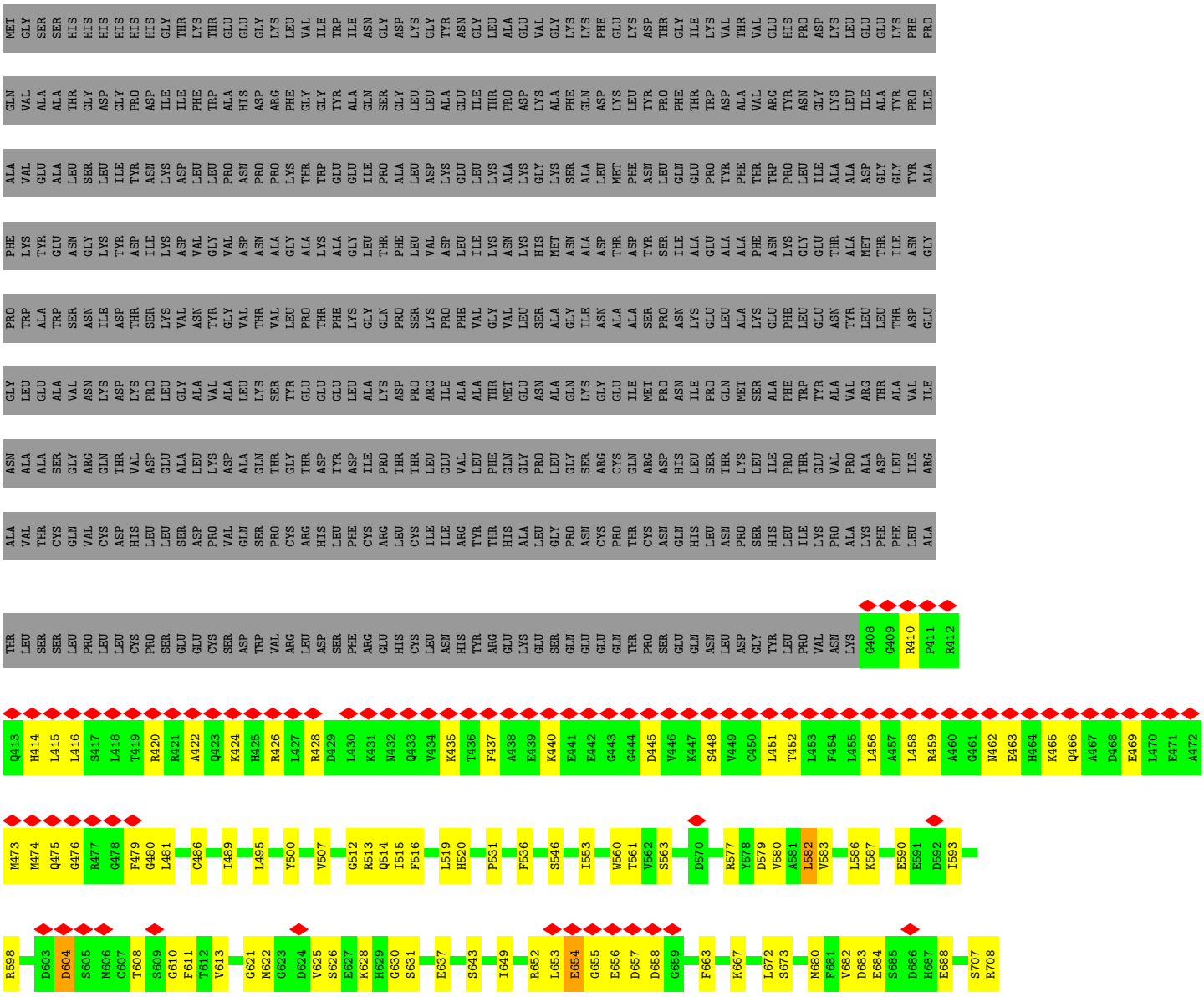
3 Residue-property plots (i)

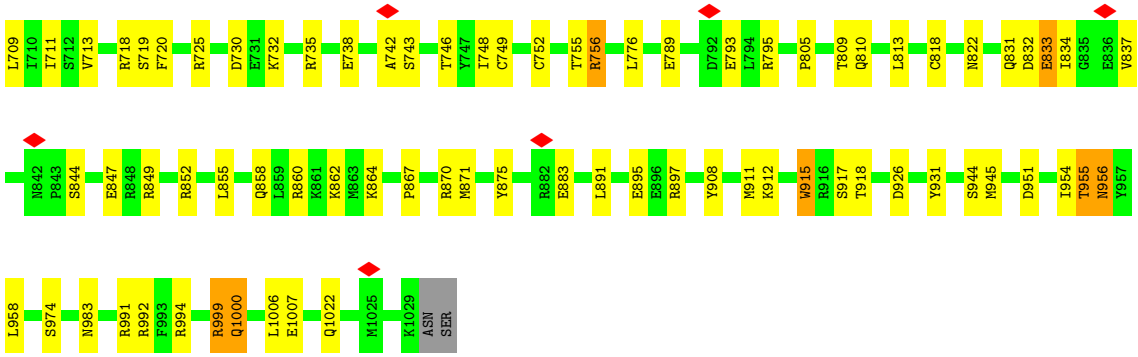
These plots are drawn for all protein, RNA, DNA and oligosaccharide chains in the entry. The first graphic for a chain summarises the proportions of the various outlier classes displayed in the second graphic. The second graphic shows the sequence view annotated by issues in geometry and atom inclusion 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: Recombination activating gene 1 - MBP chimera

Chain C:

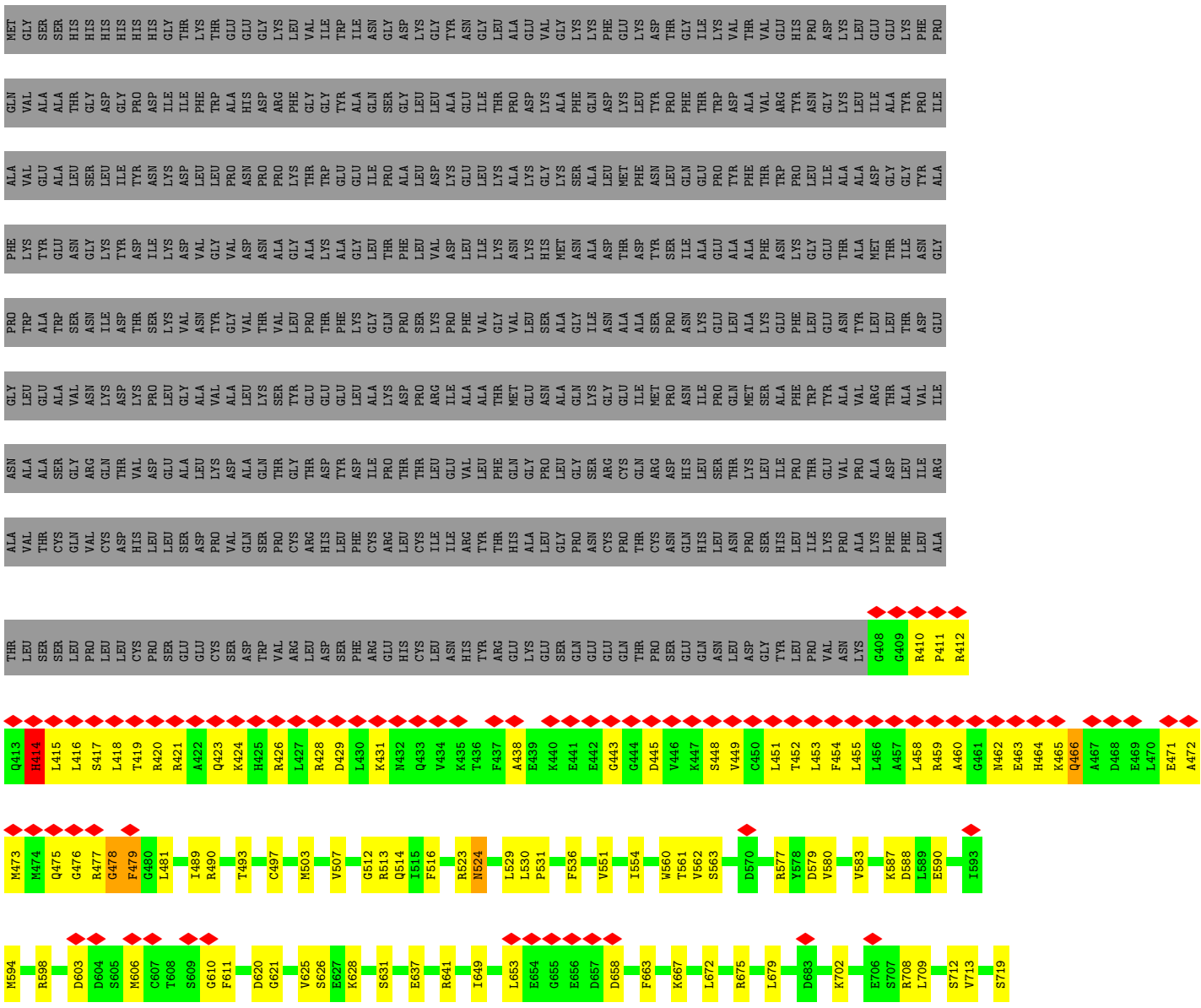


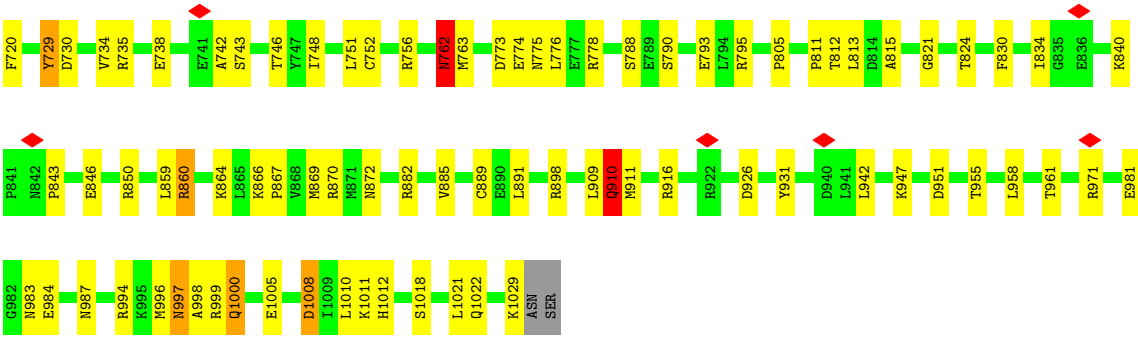




- Molecule 1: Recombination activating gene 1 - MBP chimera

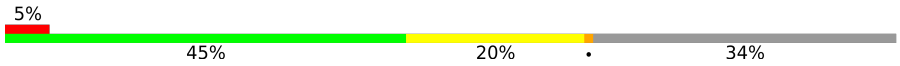
Chain A: 8% 38% 15% • 46%

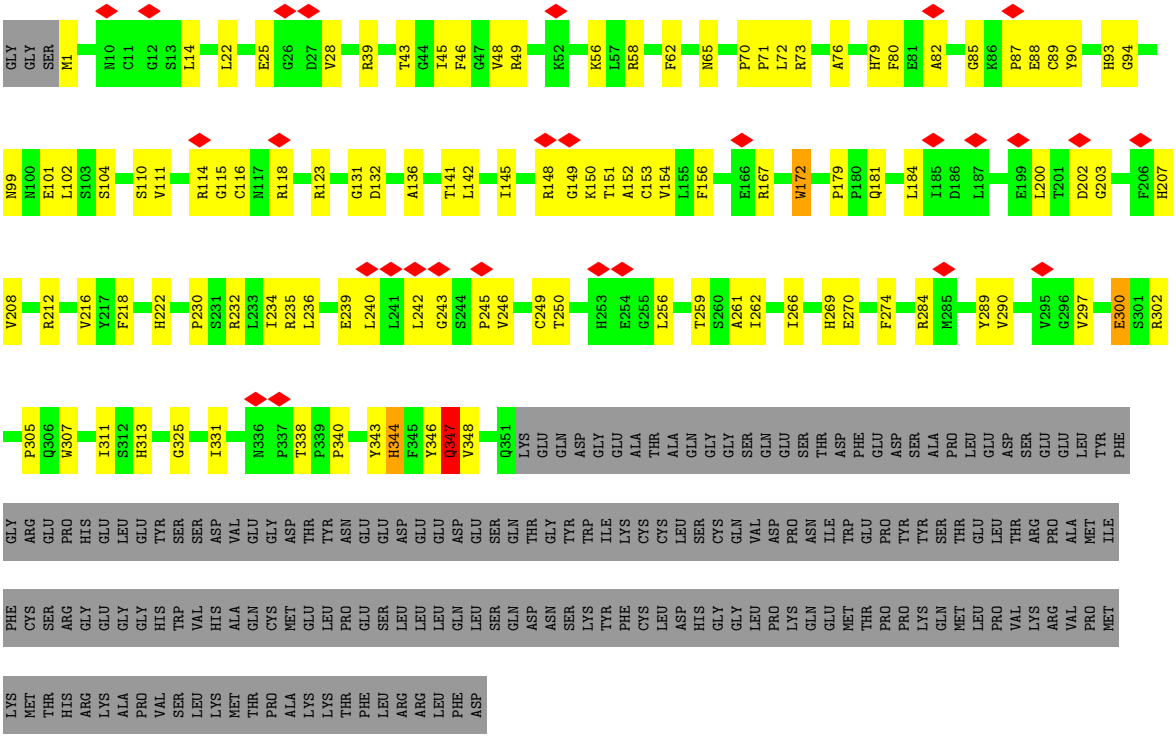




- Molecule 2: Recombination activating gene 2

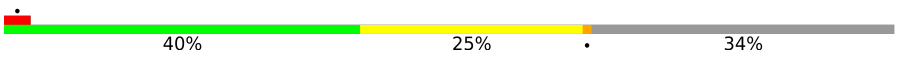
Chain D:

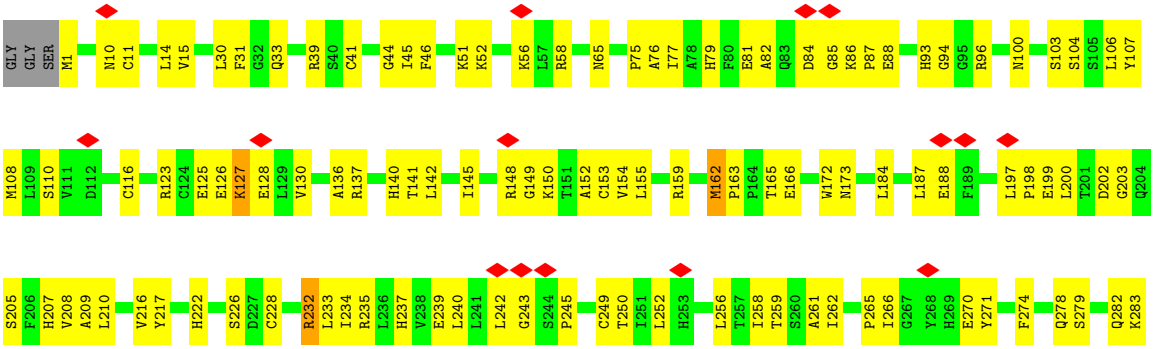


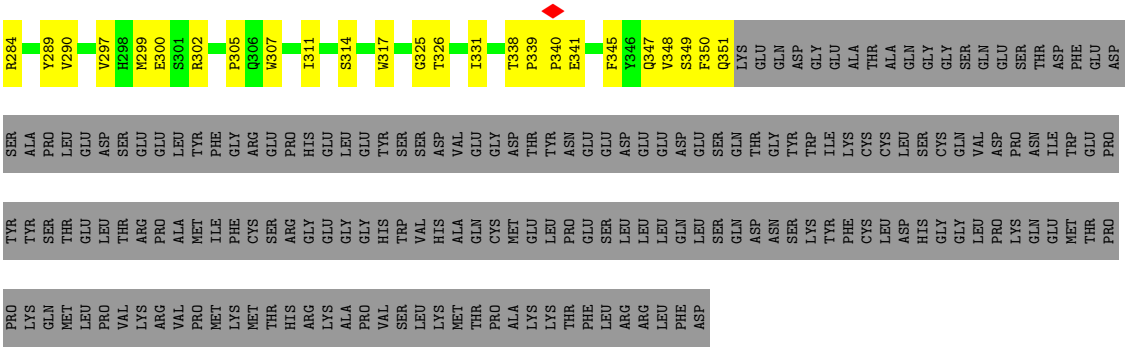


- Molecule 2: Recombination activating gene 2

Chain B:

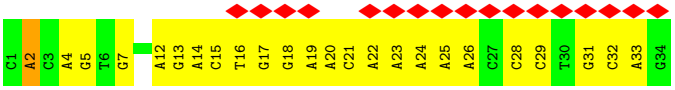






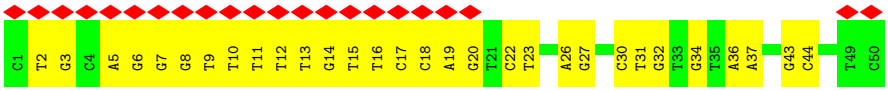
- Molecule 3: Forward strand of 12-RSS signal end

Chain E:



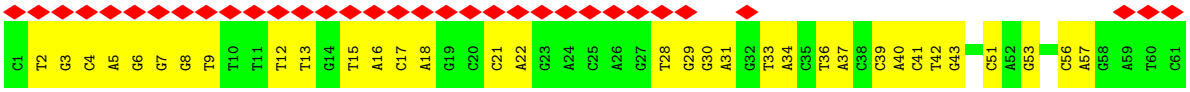
- Molecule 4: Reverse strand of 12-RSS

Chain F:



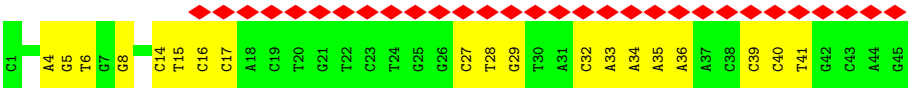
- Molecule 5: Reverse strand of 23-RSS

Chain G:



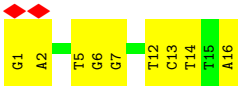
- Molecule 6: Forward strand of 23-RSS signal end

Chain H:



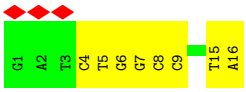
- Molecule 7: Forward strand of coding flank

Chain I:



- Molecule 7: Forward strand of coding flank

Chain J:



4 Experimental information (i)

Property	Value	Source
EM reconstruction method	SINGLE PARTICLE	Depositor
Imposed symmetry	POINT, C1	Depositor
Number of particles used	53109	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 ($e^-/Å^2$)	40	Depositor
Minimum defocus (nm)	Not provided	
Maximum defocus (nm)	Not provided	
Magnification	Not provided	
Image detector	GATAN K2 SUMMIT (4k x 4k)	Depositor
Maximum map value	0.192	Depositor
Minimum map value	-0.106	Depositor
Average map value	-0.000	Depositor
Map value standard deviation	0.008	Depositor
Recommended contour level	0.03	Depositor
Map size (Å)	271.36, 271.36, 271.36	wwPDB
Map dimensions	256, 256, 256	wwPDB
Map angles (°)	90.0, 90.0, 90.0	wwPDB
Pixel spacing (Å)	1.06, 1.06, 1.06	Depositor

5 Model quality (i)

5.1 Standard geometry (i)

Bond lengths and bond angles in the following residue types are not validated in this section: CA, ZN

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	0.60	6/5106 (0.1%)	0.87	17/6867 (0.2%)
1	C	0.38	2/5106 (0.0%)	0.65	9/6867 (0.1%)
2	B	0.38	1/2784 (0.0%)	0.77	6/3784 (0.2%)
2	D	0.52	4/2784 (0.1%)	0.81	14/3784 (0.4%)
3	E	0.70	0/784	0.90	1/1206 (0.1%)
4	F	0.77	0/1150	1.02	0/1774
5	G	0.66	0/1410	0.94	1/2175 (0.0%)
6	H	0.61	0/1030	0.92	0/1586
7	I	0.75	0/363	1.07	0/558
7	J	0.65	0/363	1.03	0/558
All	All	0.54	13/20880 (0.1%)	0.83	48/29159 (0.2%)

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	5
2	D	0	2
All	All	0	7

The worst 5 of 13 bond length outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(Å)	Ideal(Å)
1	A	910	GLN	CD-OE1	-21.19	0.77	1.24
1	A	910	GLN	CD-NE2	17.51	1.76	1.32
2	D	347	GLN	CD-OE1	-14.75	0.91	1.24
1	A	762	ASN	CB-CG	11.73	1.78	1.51
2	D	347	GLN	CD-NE2	9.09	1.55	1.32

The worst 5 of 48 bond angle outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(^{o})	Ideal(^{o})
1	A	524	ASN	CB-CG-OD1	29.19	179.98	121.60
2	B	173	ASN	CB-CG-OD1	27.53	176.65	121.60
1	A	910	GLN	CA-CB-CG	26.34	171.34	113.40
1	C	956	ASN	CB-CG-OD1	24.11	169.82	121.60
1	A	762	ASN	CB-CA-C	-21.01	68.39	110.40

There are no chirality outliers.

5 of 7 planarity outliers are listed below:

Mol	Chain	Res	Type	Group
1	A	478	GLY	Peptide
1	A	523	ARG	Peptide
1	A	762	ASN	Sidechain
2	D	344	HIS	Sidechain
2	D	347	GLN	Sidechain

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	5008	0	4967	189	0
1	C	5008	0	4967	153	0
2	B	2714	0	2665	97	0
2	D	2714	0	2664	70	0
3	E	697	0	379	32	0
4	F	1027	0	566	42	0
5	G	1256	0	686	34	0
6	H	919	0	506	23	0
7	I	326	0	182	7	0
7	J	326	0	183	10	0
8	A	1	0	0	0	0
8	C	1	0	0	0	0
9	A	2	0	0	0	0
9	C	2	0	0	0	0
All	All	20001	0	17765	565	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 15.

The worst 5 of 565 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:A:762:ASN:CB	1:A:762:ASN:CG	1.78	1.50
1:A:910:GLN:CD	1:A:910:GLN:NE2	1.76	1.37
1:C:448:SER:CB	1:A:415:LEU:CD1	2.02	1.35
1:C:448:SER:CB	1:A:415:LEU:HD11	1.58	1.32
1:A:458:LEU:HB3	1:A:462:ASN:OD1	1.41	1.19

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	620/1159 (54%)	592 (96%)	26 (4%)	2 (0%)	41	72
1	C	620/1159 (54%)	593 (96%)	27 (4%)	0	100	100
2	B	349/533 (66%)	330 (95%)	19 (5%)	0	100	100
2	D	349/533 (66%)	322 (92%)	27 (8%)	0	100	100
All	All	1938/3384 (57%)	1837 (95%)	99 (5%)	2 (0%)	54	82

All (2) Ramachandran outliers are listed below:

Mol	Chain	Res	Type
1	A	524	ASN
1	A	562	VAL

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	548/1000 (55%)	528 (96%)	20 (4%)	35	63
1	C	548/1000 (55%)	534 (97%)	14 (3%)	46	72
2	B	303/465 (65%)	297 (98%)	6 (2%)	55	77
2	D	303/465 (65%)	297 (98%)	6 (2%)	55	77
All	All	1702/2930 (58%)	1656 (97%)	46 (3%)	48	70

5 of 46 residues with a non-rotameric sidechain are listed below:

Mol	Chain	Res	Type
1	A	588	ASP
1	A	997	ASN
1	A	702	LYS
1	A	860	ARG
1	A	1000	GLN

Sometimes sidechains can be flipped to improve hydrogen bonding and reduce clashes. 5 of 11 such sidechains are listed below:

Mol	Chain	Res	Type
1	A	1022	GLN
2	B	10	ASN
2	B	237	HIS
2	B	173	ASN
1	A	762	ASN

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)

Of 6 ligands modelled in this entry, 6 are monoatomic - leaving 0 for Mogul analysis.

There are no bond length outliers.

There are no bond angle outliers.

There are no chirality outliers.

There are no torsion outliers.

There are no ring outliers.

No monomer is involved in short contacts.

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.

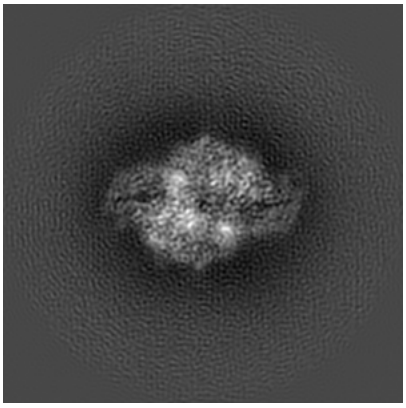
6 Map visualisation (i)

This section contains visualisations of the EMDB entry EMD-7843. These allow visual inspection of the internal detail of the map and identification of artifacts.

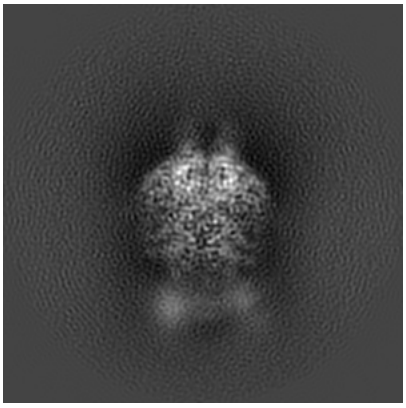
No raw map or half-maps were deposited for this entry and therefore no images, graphs, etc. pertaining to the raw map can be shown.

6.1 Orthogonal projections (i)

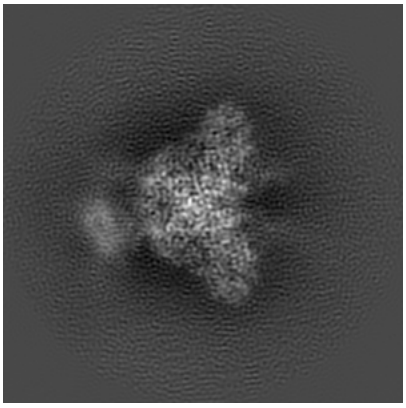
6.1.1 Primary map



X



Y

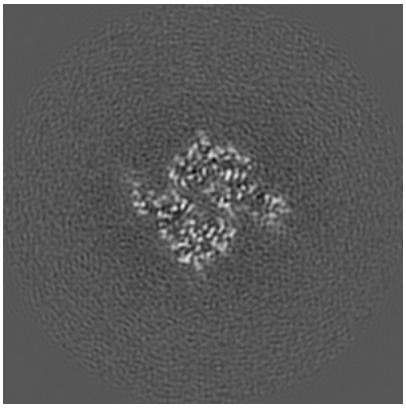


Z

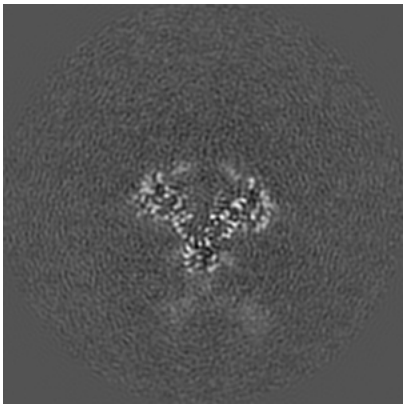
The images above show the map projected in three orthogonal directions.

6.2 Central slices (i)

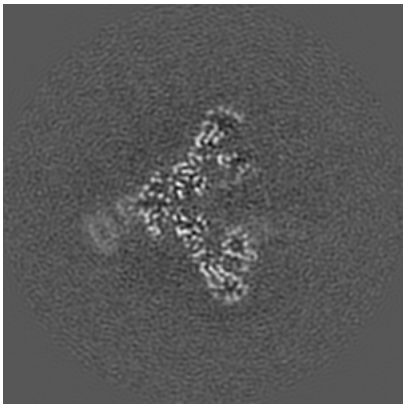
6.2.1 Primary map



X Index: 128



Y Index: 128

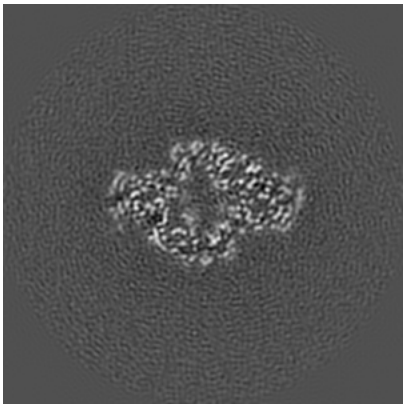


Z Index: 128

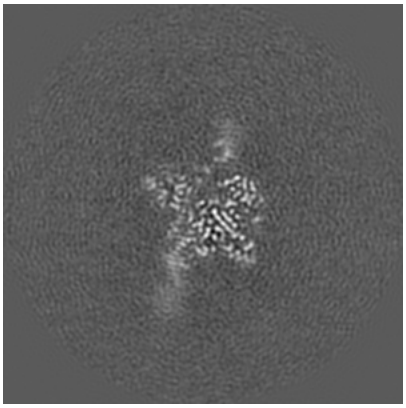
The images above show central slices of the map in three orthogonal directions.

6.3 Largest variance slices (i)

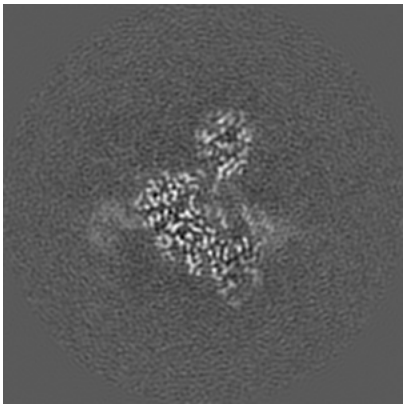
6.3.1 Primary map



X Index: 140



Y Index: 143

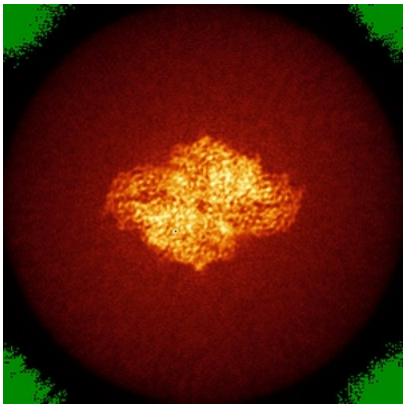


Z Index: 120

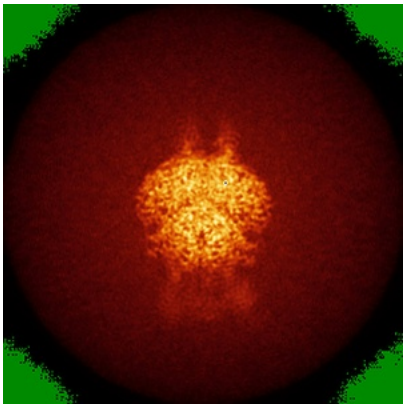
The images above show the largest variance slices of the map in three orthogonal directions.

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

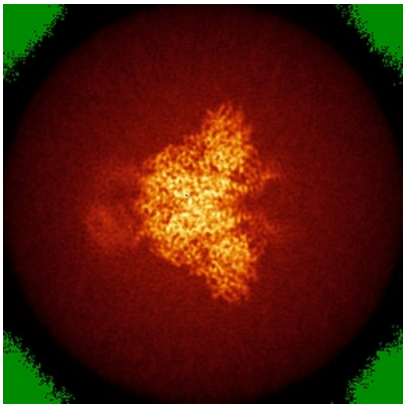
6.4.1 Primary map



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

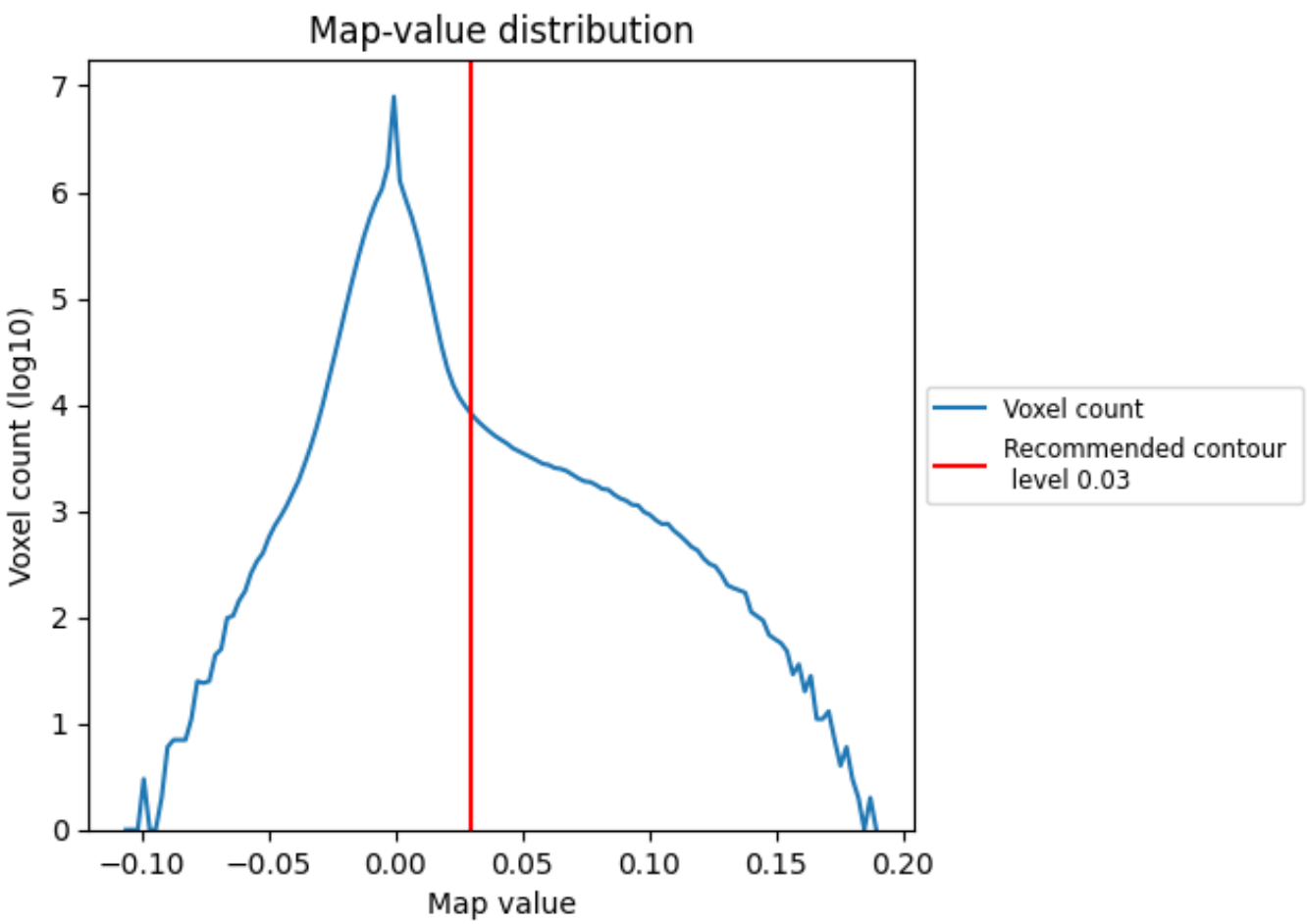
6.6 Mask visualisation (i)

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

7 Map analysis (i)

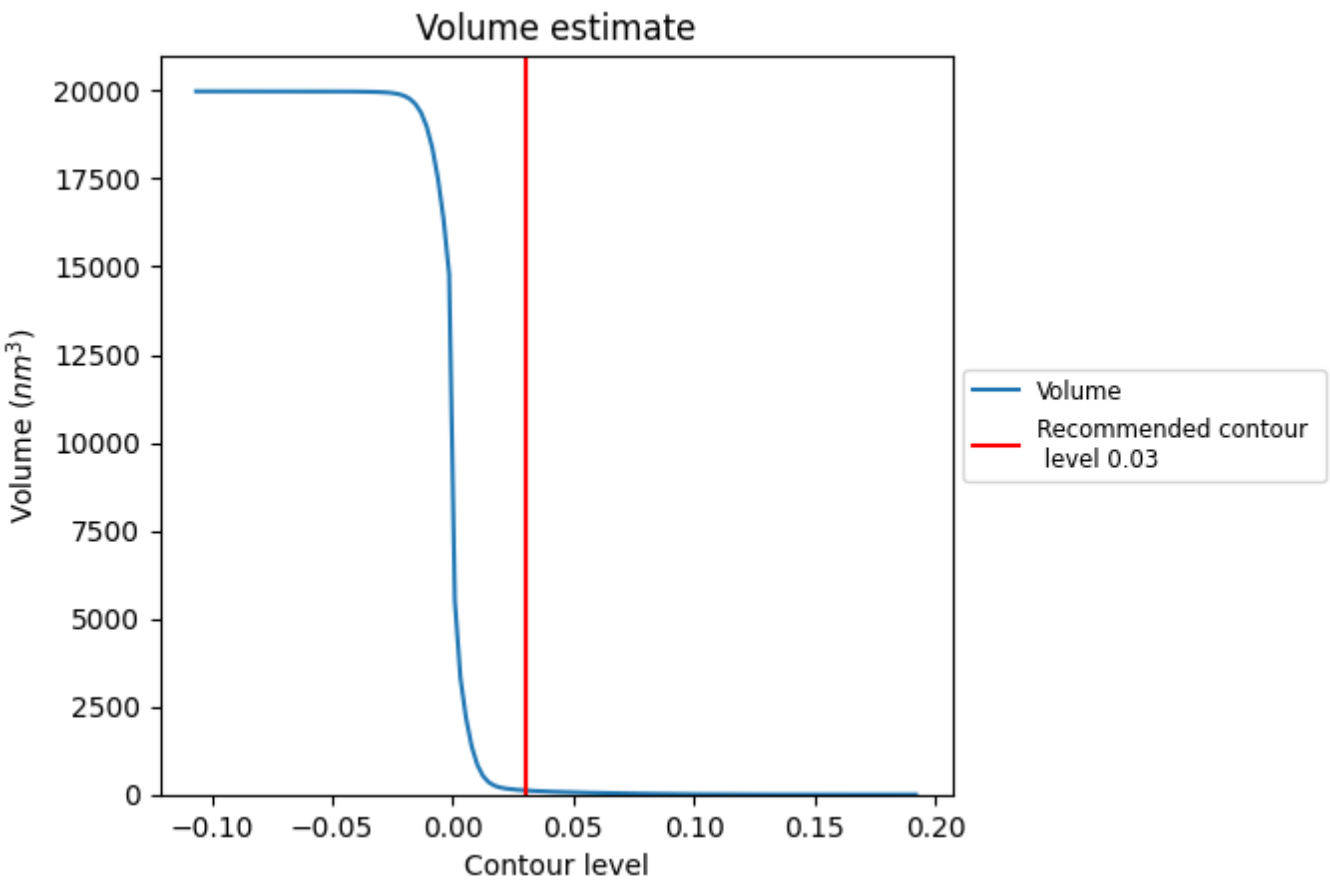
This section contains the results of statistical analysis of the map.

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.

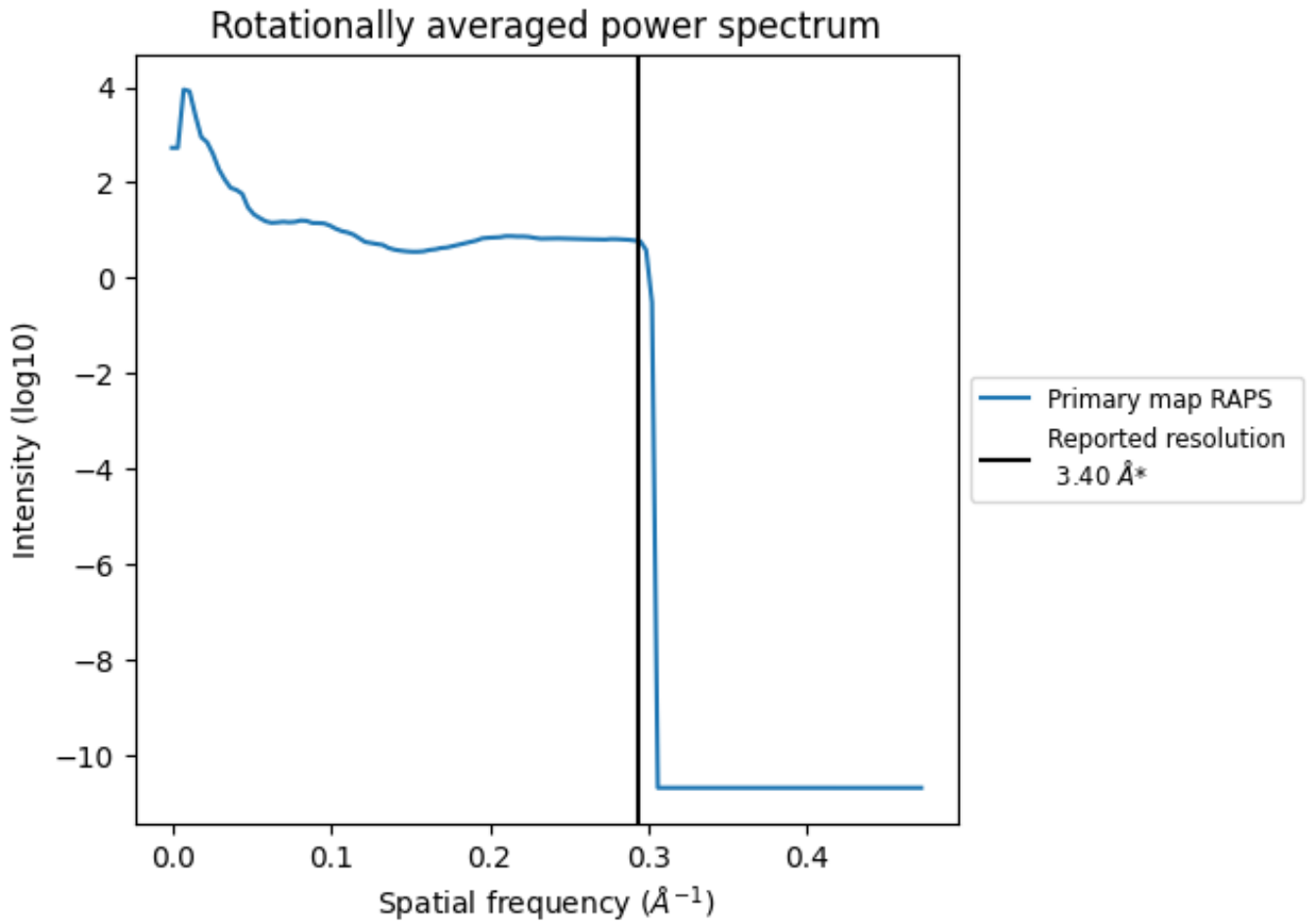
7.2 Volume estimate (i)



The volume at the recommended contour level is 119 nm^3; this corresponds to an approximate mass of 107 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 0.294 Å^{-1}

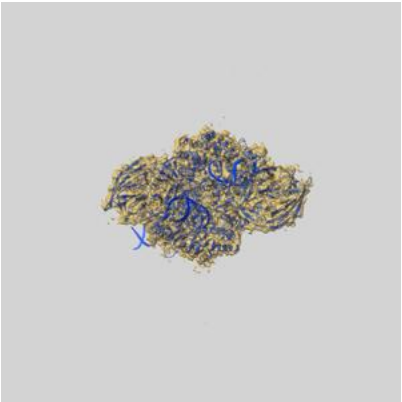
8 Fourier-Shell correlation (i)

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

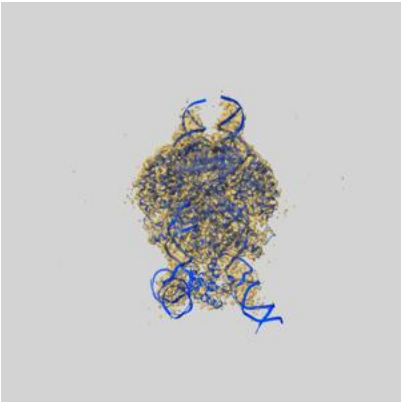
9 Map-model fit (i)

This section contains information regarding the fit between EMDB map EMD-7843 and PDB model 6DBI. Per-residue inclusion information can be found in section 3 on page 8.

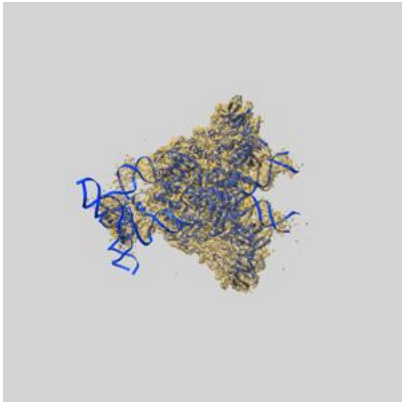
9.1 Map-model overlay (i)



X



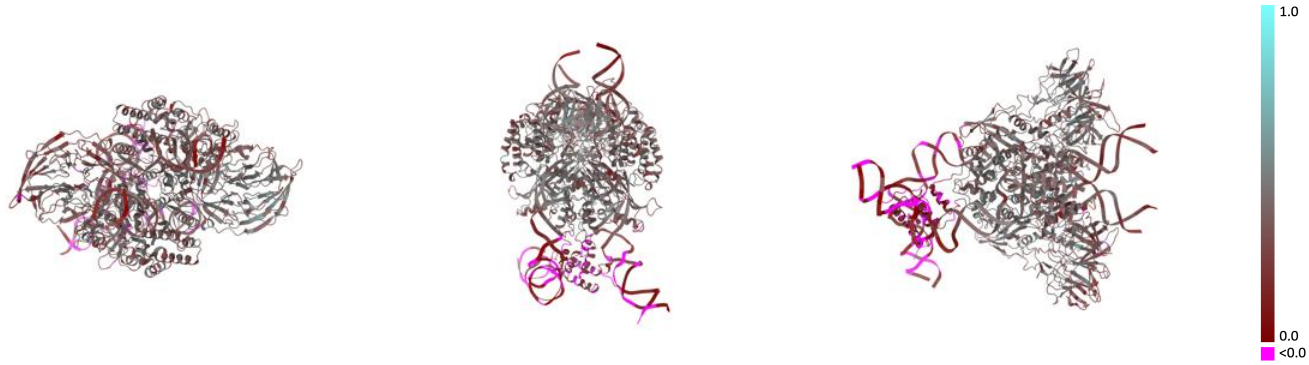
Y



Z

The images above show the 3D surface view of the map at the recommended contour level 0.03 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.

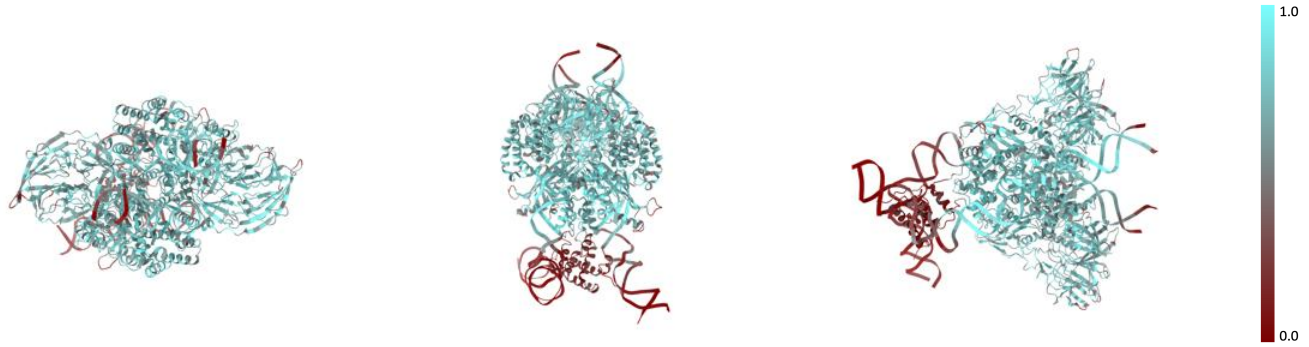
9.2 Q-score mapped to coordinate model (i)



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.

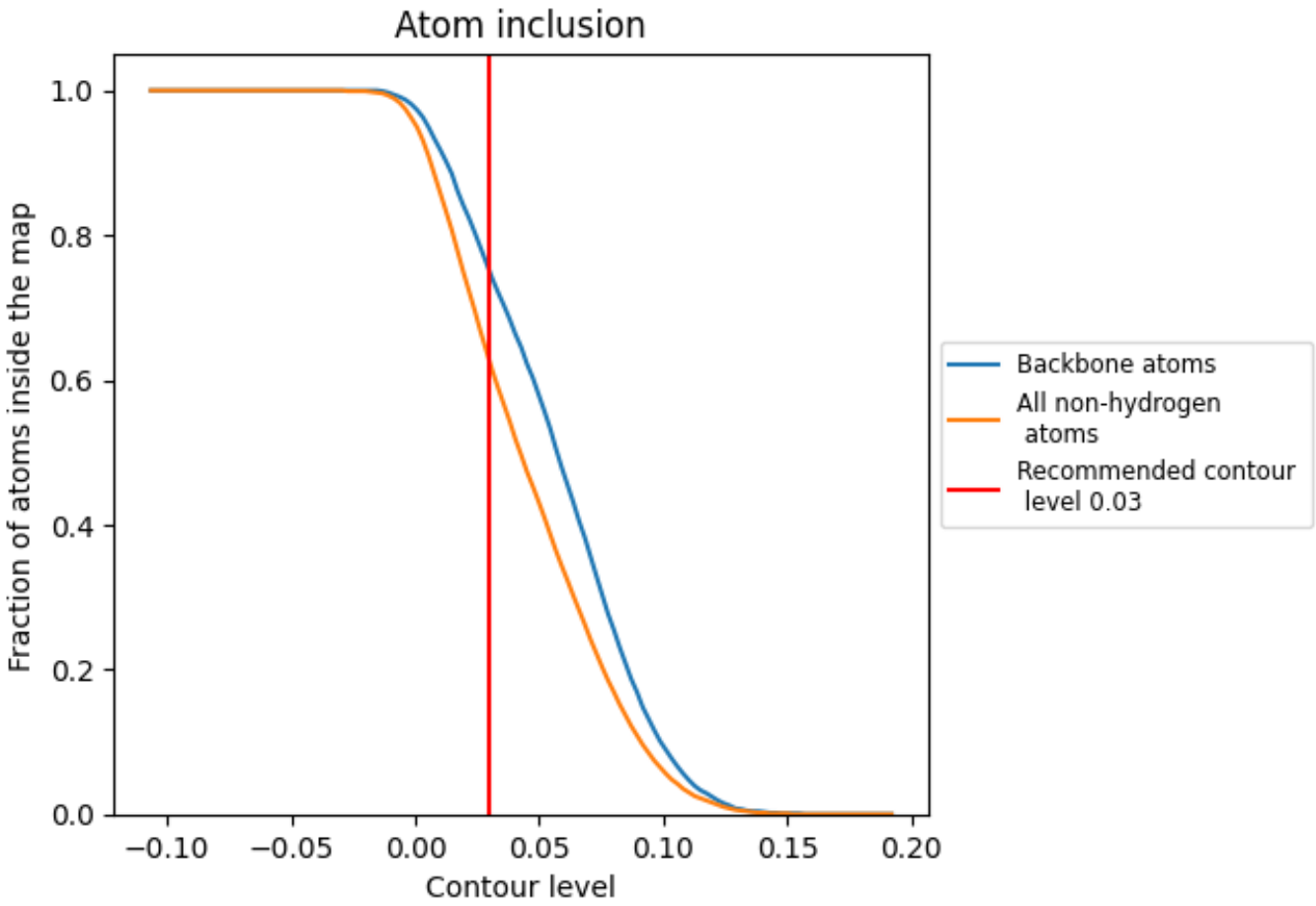
9.3 Atom inclusion mapped to coordinate model (i)



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 (0.03).

9.4 Atom inclusion (i)



At the recommended contour level, 75% of all backbone atoms, 63% 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 (0.03) and Q-score for the entire model and for each chain.

Chain	Atom inclusion	Q-score
All	0.6260	0.3240
A	0.6730	0.3530
B	0.7230	0.4060
C	0.6660	0.3490
D	0.6710	0.3460
E	0.4810	0.1900
F	0.5240	0.2220
G	0.3790	0.1930
H	0.2980	0.1430
I	0.6960	0.2500
J	0.5950	0.2840