Full wwPDB NMR Structure Validation Report (i)

Jun 4, 2023 – 11:02 AM EDT

PDB ID	:	2N1B
BMRB ID	:	25554
Title	:	NMR solution structure of nucleotide-free Ran GTPase
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Deposited on	:	2015-03-26

This is a Full wwPDB NMR Structure Validation 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/NMRValidationReportHelp
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:

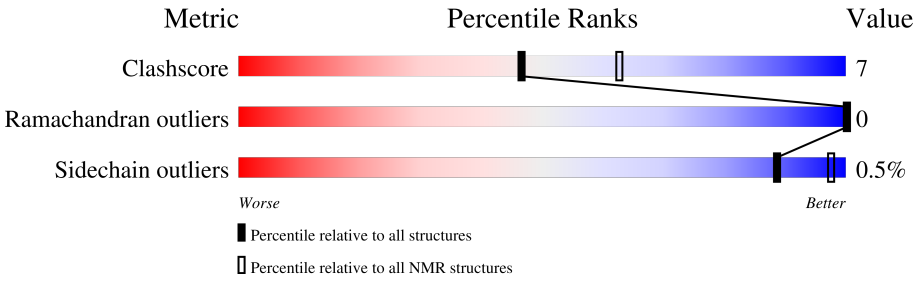
MolProbity	:	4.02b-467
Percentile statistics	:	20191225.v01 (using entries in the PDB archive December 25th 2019)
wwPDB-RCI	:	v_1n_11_5_13_A (Berjanski et al., 2005)
PANAV	:	Wang et al. (2010)
wwPDB-ShiftChecker	:	v1.2
BMRB Restraints Analysis	:	v1.2
Ideal geometry (proteins)	:	Engh & Huber (2001)
Ideal geometry (DNA, RNA)	:	Parkinson et al. (1996)
Validation Pipeline (wwPDB-VP)	:	2.33

1 Overall quality at a glance

The following experimental techniques were used to determine the structure:
SOLUTION NMR

The overall completeness of chemical shifts assignment is 83%.

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)	NMR archive (#Entries)
Clashscore	158937	12864
Ramachandran outliers	154571	11451
Sidechain outliers	154315	11428

The table below summarises the geometric issues observed across the polymeric chains and their fit to the experimental data. The red, orange, yellow and green segments indicate the fraction of residues that contain outliers for >=3, 2, 1 and 0 types of geometric quality criteria. A cyan segment indicates the fraction of residues that are not part of the well-defined cores, and 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%

Mol	Chain	Length	Quality of chain
1	A	261	67% 14% • 17%

2 Ensemble composition and analysis

This entry contains 10 models. Model 2 is the overall representative, medoid model (most similar to other models). The authors have identified model 1 as representative, based on the following criterion: *fewest violations*.

The following residues are included in the computation of the global validation metrics.

Well-defined (core) protein residues			
Well-defined core	**Residue range (total)**	**Backbone RMSD (Å)**	**Medoid model**
1	A:5-A:216 (212)	0.04	2

Ill-defined regions of proteins are excluded from the global statistics.

Ligands and non-protein polymers are included in the analysis.

The models can be grouped into 2 clusters and 1 single-model cluster was found.

Cluster number	Models
1	1, 2, 4, 7, 8
2	3, 5, 9, 10
Single-model clusters	6

3 Entry composition (i)

There is only 1 type of molecule in this entry. The entry contains 3447 atoms, of which 1724 are hydrogens and 0 are deuteriums.

- Molecule 1 is a protein called GTP-binding nuclear protein Ran.

Mol	Chain	Residues	Atoms						Trace
			Total	C	H	N	O	S	
1	A	216	3447	1109	1724	295	312	7	0

There are 45 discrepancies between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	Comment	Reference
A	-40	MET	-	expression tag	UNP P62826
A	-39	ARG	-	expression tag	UNP P62826
A	-38	GLY	-	expression tag	UNP P62826
A	-37	SER	-	expression tag	UNP P62826
A	-36	HIS	-	expression tag	UNP P62826
A	-35	HIS	-	expression tag	UNP P62826
A	-34	HIS	-	expression tag	UNP P62826
A	-33	HIS	-	expression tag	UNP P62826
A	-32	HIS	-	expression tag	UNP P62826
A	-31	HIS	-	expression tag	UNP P62826
A	-30	GLY	-	expression tag	UNP P62826
A	-29	MET	-	expression tag	UNP P62826
A	-28	ALA	-	expression tag	UNP P62826
A	-27	SER	-	expression tag	UNP P62826
A	-26	MET	-	expression tag	UNP P62826
A	-25	THR	-	expression tag	UNP P62826
A	-24	GLY	-	expression tag	UNP P62826
A	-23	GLY	-	expression tag	UNP P62826
A	-22	GLN	-	expression tag	UNP P62826
A	-21	GLN	-	expression tag	UNP P62826
A	-20	MET	-	expression tag	UNP P62826
A	-19	GLY	-	expression tag	UNP P62826
A	-18	ARG	-	expression tag	UNP P62826
A	-17	ASP	-	expression tag	UNP P62826
A	-16	LEU	-	expression tag	UNP P62826
A	-15	TYR	-	expression tag	UNP P62826
A	-14	ASP	-	expression tag	UNP P62826
A	-13	ASP	-	expression tag	UNP P62826
A	-12	ASP	-	expression tag	UNP P62826
A	-11	ASP	-	expression tag	UNP P62826
A	-10	LYS	-	expression tag	UNP P62826

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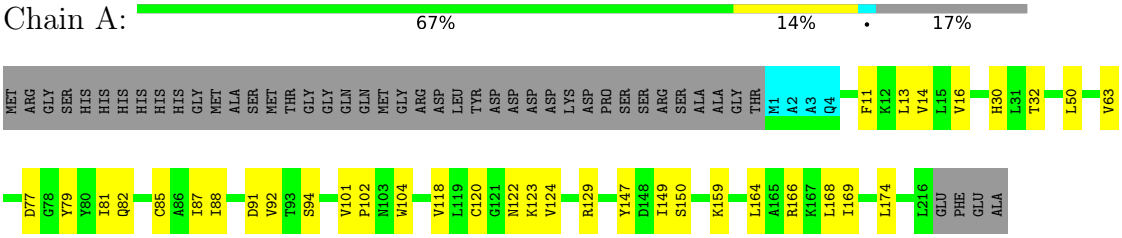
Chain	Residue	Modelled	Actual	Comment	Reference
A	-9	ASP	-	expression tag	UNP P62826
A	-8	PRO	-	expression tag	UNP P62826
A	-7	SER	-	expression tag	UNP P62826
A	-6	SER	-	expression tag	UNP P62826
A	-5	ARG	-	expression tag	UNP P62826
A	-4	SER	-	expression tag	UNP P62826
A	-3	ALA	-	expression tag	UNP P62826
A	-2	ALA	-	expression tag	UNP P62826
A	-1	GLY	-	expression tag	UNP P62826
A	0	THR	-	expression tag	UNP P62826
A	217	GLU	-	expression tag	UNP P62826
A	218	PHE	-	expression tag	UNP P62826
A	219	GLU	-	expression tag	UNP P62826
A	220	ALA	-	expression tag	UNP P62826

4 Residue-property plots

4.1 Average score per residue in the NMR ensemble

These plots are provided for all protein, RNA, DNA and oligosaccharide chains in the entry. The first graphic is the same as shown in the summary in section 1 of this report. The second graphic shows the sequence where residues are colour-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. Stretches of 2 or more consecutive residues without any outliers are shown as green connectors. Residues which are classified as ill-defined in the NMR ensemble, are shown in cyan with an underline colour-coded according to the previous scheme. Residues which were present in the experimental sample, but not modelled in the final structure are shown in grey.

• Molecule 1: GTP-binding nuclear protein Ran

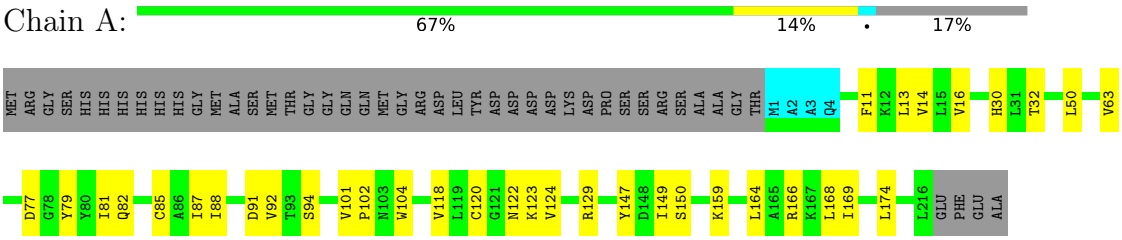


4.2 Scores per residue for each member of the ensemble

Colouring as in section 4.1 above.

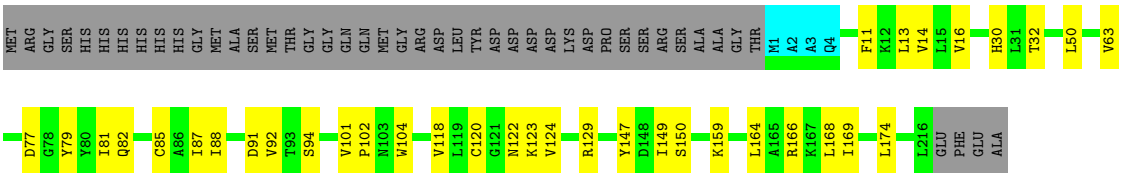
4.2.1 Score per residue for model 1

• Molecule 1: GTP-binding nuclear protein Ran



4.2.2 Score per residue for model 2 (medoid)

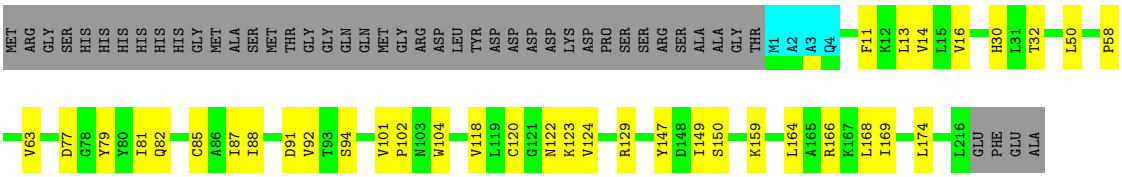
• Molecule 1: GTP-binding nuclear protein Ran



4.2.3 Score per residue for model 3

• Molecule 1: GTP-binding nuclear protein Ran

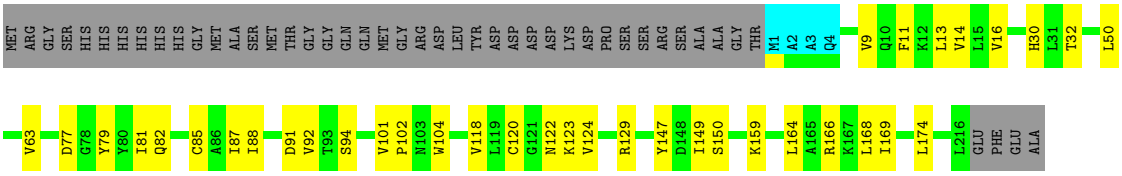
Chain A:



4.2.4 Score per residue for model 4

• Molecule 1: GTP-binding nuclear protein Ran

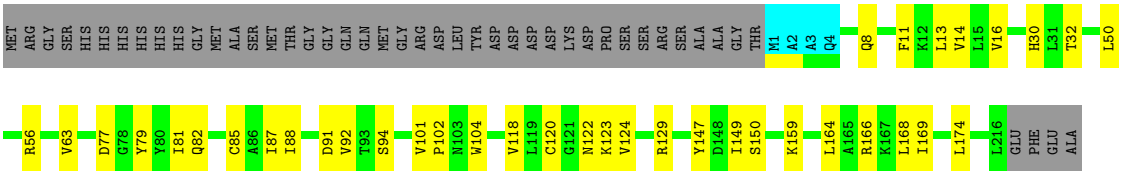
Chain A:



4.2.5 Score per residue for model 5

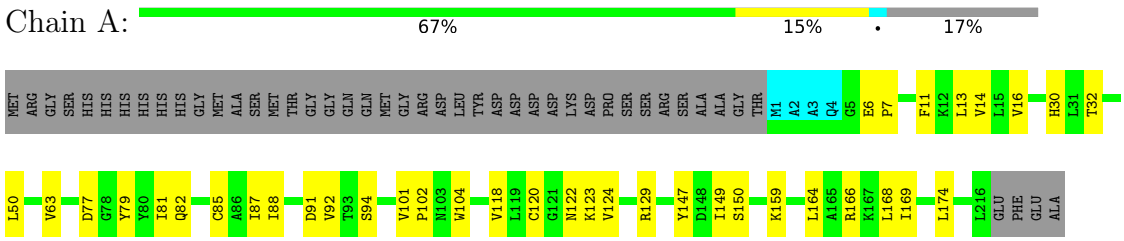
• Molecule 1: GTP-binding nuclear protein Ran

Chain A:



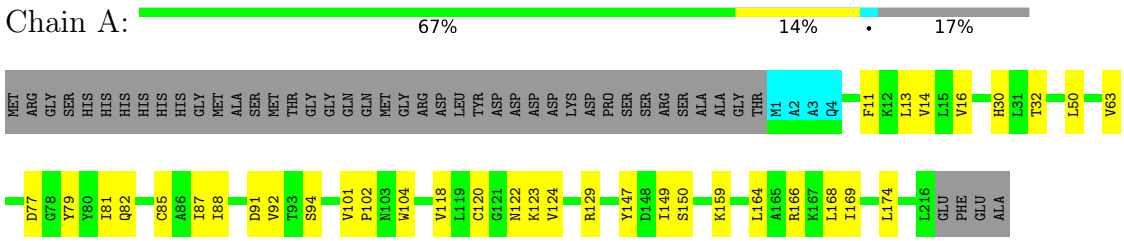
4.2.6 Score per residue for model 6

• Molecule 1: GTP-binding nuclear protein Ran



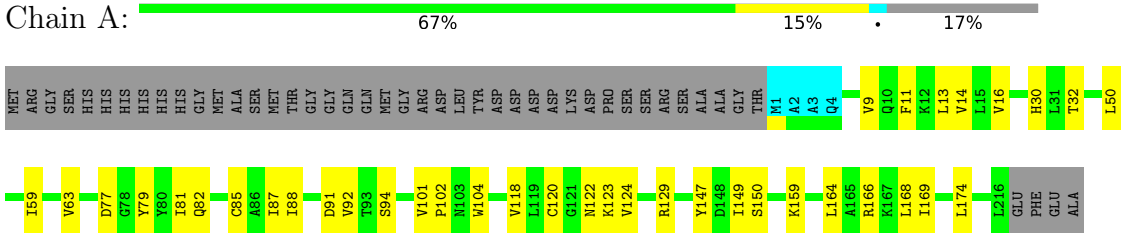
4.2.7 Score per residue for model 7

• Molecule 1: GTP-binding nuclear protein Ran



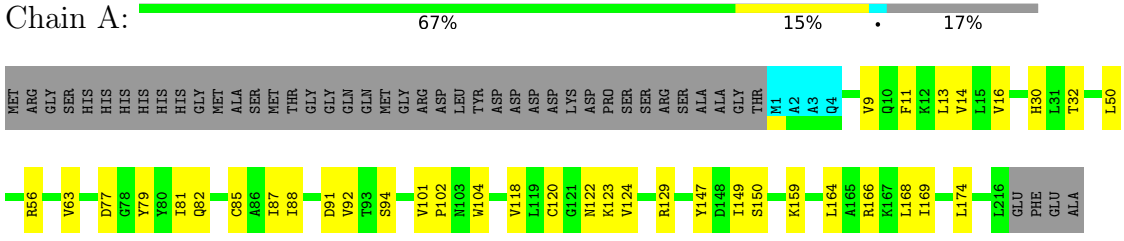
4.2.8 Score per residue for model 8

• Molecule 1: GTP-binding nuclear protein Ran



4.2.9 Score per residue for model 9

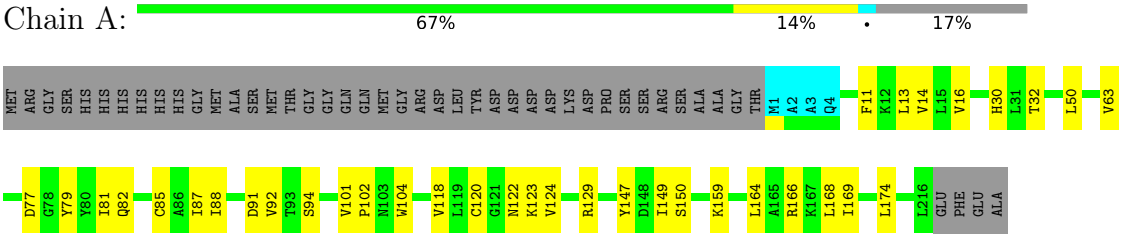
• Molecule 1: GTP-binding nuclear protein Ran



4.2.10 Score per residue for model 10

• Molecule 1: GTP-binding nuclear protein Ran

Chain A:



5 Refinement protocol and experimental data overview

The models were refined using the following method: *torsion angle dynamics*.

Of the 50 calculated structures, 10 were deposited, based on the following criterion: *structures with the least restraint violations*.

The following table shows the software used for structure solution, optimisation and refinement.

Software name	Classification	Version
CYANA	refinement	

The following table shows chemical shift validation statistics as aggregates over all chemical shift files. Detailed validation can be found in section 7 of this report.

Chemical shift file(s)	working_cs.cif
Number of chemical shift lists	1
Total number of shifts	2589
Number of shifts mapped to atoms	2584
Number of unparsed shifts	0
Number of shifts with mapping errors	5
Number of shifts with mapping warnings	0
Assignment completeness (well-defined parts)	83%

6 Model quality

6.1 Standard geometry

There are no covalent bond-length or bond-angle outliers.

There are no bond-length outliers.

There are no bond-angle outliers.

There are no chirality outliers.

There are no planarity outliers.

6.2 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 each 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 averaged over the ensemble.

Mol	Chain	Non-H	H(model)	H(added)	Clashes
1	A	1696	1697	1696	22±0
All	All	16960	16970	16960	221

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.

All unique clashes are listed below, sorted by their clash magnitude.

Atom-1	Atom-2	Clash(Å)	Distance(Å)	Models	
				Worst	Total
1:A:120:CYS:SG	1:A:147:TYR:HB2	0.76	2.21	1	10
1:A:124:VAL:HG12	1:A:150:SER:HB2	0.62	1.71	1	10
1:A:87:ILE:HG12	1:A:118:VAL:HB	0.60	1.73	1	10
1:A:6:GLU:HB3	1:A:7:PRO:HD2	0.60	1.74	6	1
1:A:11:PHE:HB3	1:A:85:CYS:SG	0.57	2.40	1	10
1:A:101:VAL:HB	1:A:102:PRO:HD3	0.55	1.79	1	10
1:A:14:VAL:HG13	1:A:79:TYR:CE2	0.54	2.38	1	10
1:A:118:VAL:HG13	1:A:147:TYR:CE1	0.50	2.40	1	10
1:A:92:VAL:HB	1:A:129:ARG:HG3	0.50	1.82	1	10
1:A:147:TYR:HD2	1:A:159:LYS:HE2	0.50	1.65	1	10
1:A:30:HIS:CE1	1:A:32:THR:HB	0.49	2.43	1	10
1:A:122:ASN:O	1:A:123:LYS:HB2	0.49	2.07	1	10
1:A:13:LEU:HD23	1:A:63:VAL:HG22	0.48	1.84	1	10
1:A:30:HIS:CG	1:A:50:LEU:HD22	0.47	2.45	1	10

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Atom-1	Atom-2	Clash(Å)	Distance(Å)	Models	
				Worst	Total
1:A:169:ILE:HD12	1:A:174:LEU:HD22	0.46	1.87	1	10
1:A:85:CYS:HB3	1:A:164:LEU:HD21	0.45	1.86	1	10
1:A:122:ASN:HA	1:A:149:ILE:O	0.45	2.11	1	10
1:A:16:VAL:HG11	1:A:104:TRP:HB3	0.44	1.89	1	10
1:A:81:ILE:HG22	1:A:82:GLN:HG3	0.42	1.92	1	10
1:A:166:ARG:HG2	1:A:174:LEU:HB3	0.41	1.91	1	10
1:A:85:CYS:SG	1:A:168:LEU:HD11	0.41	2.55	1	10
1:A:16:VAL:HB	1:A:88:ILE:HG12	0.40	1.92	1	10
1:A:91:ASP:HB3	1:A:94:SER:HB3	0.40	1.94	1	10

6.3 Torsion angles (i)

6.3.1 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 NMR 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	211/261 (81%)	209±0 (99±0%)	2±0 (1±0%)	0±0 (0±0%)	100	100
All	All	2110/2610 (81%)	2087 (99%)	23 (1%)	0 (0%)	100	100

There are no Ramachandran outliers.

6.3.2 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 NMR 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	183/220 (83%)	182±0 (99±0%)	1±0 (1±0%)	89	97
All	All	1830/2200 (83%)	1820 (99%)	10 (1%)	89	97

All 1 unique residues with a non-rotameric sidechain are listed below.

Mol	Chain	Res	Type	Models (Total)
1	A	77	ASP	10

6.3.3 RNA

There are no RNA molecules in this entry.

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

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

6.5 Carbohydrates

There are no monosaccharides in this entry.

6.6 Ligand geometry

There are no ligands in this entry.

6.7 Other polymers

There are no such molecules in this entry.

6.8 Polymer linkage issues

There are no chain breaks in this entry.

7 Chemical shift validation

The completeness of assignment taking into account all chemical shift lists is 83% for the well-defined parts and 83% for the entire structure.

7.1 Chemical shift list 1

File name: working_cs.cif

Chemical shift list name: *assigned_chem_shift_list_1*

7.1.1 Bookkeeping

The following table shows the results of parsing the chemical shift list and reports the number of nuclei with statistically unusual chemical shifts.

Total number of shifts	2589
Number of shifts mapped to atoms	2584
Number of unparsed shifts	0
Number of shifts with mapping errors	5
Number of shifts with mapping warnings	0
Number of shift outliers (ShiftChecker)	56

The following assigned chemical shifts were not mapped to the molecules present in the coordinate file.

- No matching atom found in the structure. All 5 occurrences are reported below.

List ID	Chain	Res	Type	Atom	Shift Data		
					Value	Uncertainty	Ambiguity
1	A	30	HIS	HD1	10.269	0.020	1
1	A	53	HIS	HD1	10.367	0.020	1
1	A	105	HIS	HE2	11.112	0.020	1
1	A	139	HIS	HD1	10.687	0.020	1
1	A	199	HIS	HD1	10.494	0.020	1

7.1.2 Chemical shift referencing

The following table shows the suggested chemical shift referencing corrections.

Nucleus	# values	Correction ± precision, *ppm*	Suggested action
$^{13}C_{\alpha}$	212	-0.94 ± 0.17	Should be checked
$^{13}C_{\beta}$	198	0.15 ± 0.26	None needed (< 0.5 ppm)
$^{13}C'$	209	-0.47 ± 0.14	None needed (< 0.5 ppm)

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Nucleus	# values	Correction ± precision, *ppm*	Suggested action
^{15}N	215	1.53 ± 0.66	Should be applied

7.1.3 Completeness of resonance assignments

The following table shows the completeness of the chemical shift assignments for the well-defined regions of the structure. The overall completeness is 83%, i.e. 2463 atoms were assigned a chemical shift out of a possible 2959. 0 out of 40 assigned methyl groups (LEU and VAL) were assigned stereospecifically.

	Total	^{1}H	^{13}C	^{15}N
Backbone	839/1050 (80%)	227/426 (53%)	413/424 (97%)	199/200 (100%)
Sidechain	1363/1643 (83%)	972/1067 (91%)	328/513 (64%)	63/63 (100%)
Aromatic	261/266 (98%)	129/129 (100%)	117/122 (96%)	15/15 (100%)
Overall	2463/2959 (83%)	1328/1622 (82%)	858/1059 (81%)	277/278 (100%)

The following table shows the completeness of the chemical shift assignments for the full structure. The overall completeness is 83%, i.e. 2500 atoms were assigned a chemical shift out of a possible 3007. 0 out of 40 assigned methyl groups (LEU and VAL) were assigned stereospecifically.

	Total	^{1}H	^{13}C	^{15}N
Backbone	855/1070 (80%)	231/434 (53%)	421/432 (97%)	203/204 (100%)
Sidechain	1384/1671 (83%)	988/1086 (91%)	332/521 (64%)	64/64 (100%)
Aromatic	261/266 (98%)	129/129 (100%)	117/122 (96%)	15/15 (100%)
Overall	2500/3007 (83%)	1348/1649 (82%)	870/1075 (81%)	282/283 (100%)

7.1.4 Statistically unusual chemical shifts

The following table lists the statistically unusual chemical shifts. These are statistical measures, and large deviations from the mean do not necessarily imply incorrect assignments. Molecules containing paramagnetic centres or hemes are expected to give rise to anomalous chemical shifts.

List Id	Chain	Res	Type	Atom	Shift, *ppm*	Expected range, *ppm*	Z-score
1	A	114	ASN	ND2	1114.29	101.55 – 123.95	447.1
1	A	21	THR	HG1	6.38	0.08 – 2.19	24.8
1	A	66	THR	HG1	6.01	0.08 – 2.19	23.1
1	A	207	THR	HG1	5.99	0.08 – 2.19	23.0
1	A	206	THR	HG1	5.96	0.08 – 2.19	22.8
1	A	24	THR	HG1	5.91	0.08 – 2.19	22.6
1	A	93	THR	HG1	5.76	0.08 – 2.19	21.9
1	A	32	THR	HG1	5.71	0.08 – 2.19	21.7
1	A	54	THR	HG1	5.59	0.08 – 2.19	21.1

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List Id	Chain	Res	Type	Atom	Shift, *ppm*	Expected range, *ppm*	Z-score
1	A	97	THR	HG1	5.48	0.08 – 2.19	20.6
1	A	25	THR	HG1	5.39	0.08 – 2.19	20.1
1	A	42	THR	HG1	5.31	0.08 – 2.19	19.8
1	A	134	LYS	CB	54.58	24.03 – 41.47	12.5
1	A	175	GLU	CA	76.25	47.03 – 67.62	9.2
1	A	45	VAL	CB	17.10	23.86 – 41.50	-8.8
1	A	68	GLY	C	189.53	164.92 – 182.89	8.7
1	A	151	ALA	C	160.74	167.61 – 188.05	-8.4
1	A	183	ALA	C	160.74	167.61 – 188.05	-8.4
1	A	37	LYS	NZ	53.20	19.79 – 46.09	7.7
1	A	38	LYS	NZ	52.40	19.79 – 46.09	7.4
1	A	143	ASN	ND2	129.29	101.55 – 123.95	7.4
1	A	158	GLU	CA	72.15	47.03 – 67.62	7.2
1	A	44	GLY	CA	54.58	38.93 – 51.79	7.2
1	A	29	ARG	CA	72.15	45.44 – 68.13	6.8
1	A	33	GLY	C	161.91	164.92 – 182.89	-6.7
1	A	123	LYS	CA	70.98	46.18 – 67.77	6.5
1	A	170	GLY	C	162.50	164.92 – 182.89	-6.3
1	A	132	LYS	NZ	49.59	19.79 – 46.09	6.3
1	A	94	SER	C	163.97	166.15 – 183.14	-6.3
1	A	123	LYS	NZ	49.29	19.79 – 46.09	6.2
1	A	134	LYS	NZ	49.30	19.79 – 46.09	6.2
1	A	12	LYS	NZ	49.23	19.79 – 46.09	6.2
1	A	141	LYS	NZ	49.24	19.79 – 46.09	6.2
1	A	142	LYS	NZ	49.22	19.79 – 46.09	6.2
1	A	152	LYS	NZ	49.22	19.79 – 46.09	6.2
1	A	71	LYS	NZ	49.00	19.79 – 46.09	6.1
1	A	119	LEU	CA	42.87	45.17 – 66.21	-6.1
1	A	22	GLY	C	184.81	164.92 – 182.89	6.1
1	A	161	PHE	CE2	137.89	124.80 – 136.72	6.0
1	A	165	ALA	C	165.72	167.61 – 188.05	-5.9
1	A	110	ARG	NE	94.02	76.53 – 92.65	5.8
1	A	60	LYS	NZ	48.29	19.79 – 46.09	5.8
1	A	113	GLU	CA	69.22	47.03 – 67.62	5.8
1	A	154	ASN	ND2	125.21	101.55 – 123.95	5.6
1	A	20	GLY	N	129.37	91.59 – 127.52	5.5
1	A	127	LYS	NZ	47.33	19.79 – 46.09	5.5
1	A	29	ARG	NE	93.29	76.53 – 92.65	5.4
1	A	76	ARG	NE	93.29	76.53 – 92.65	5.4
1	A	67	ALA	N	104.86	106.13 – 140.55	-5.4
1	A	194	ALA	C	166.90	167.61 – 188.05	-5.3
1	A	167	LYS	NZ	46.77	19.79 – 46.09	5.3

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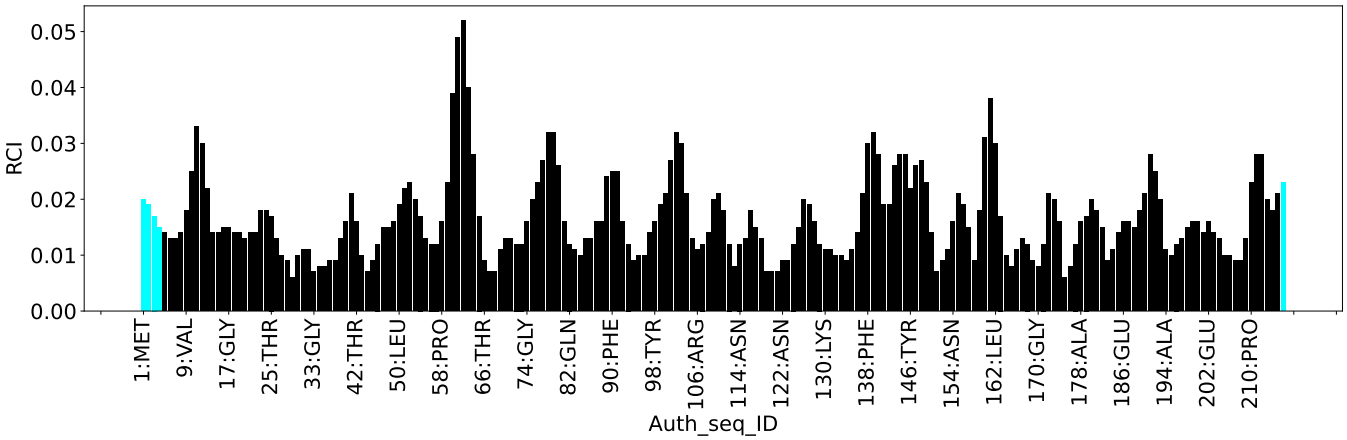
List Id	Chain	Res	Type	Atom	Shift, *ppm*	Expected range, *ppm*	Z-score
1	A	84	GLN	H	11.17	5.39 – 11.05	5.2
1	A	17	GLY	N	127.91	91.59 – 127.52	5.1
1	A	33	GLY	N	127.84	91.59 – 127.52	5.1
1	A	57	GLY	N	127.71	91.59 – 127.52	5.0
1	A	161	PHE	CE1	137.30	124.17 – 137.29	5.0

7.1.5 Random Coil Index (RCI) plots (i)

The image below reports *random coil index* values for the protein chains in the structure. The height of each bar gives a probability of a given residue to be disordered, as predicted from the available chemical shifts and the amino acid sequence. A value above 0.2 is an indication of significant predicted disorder. The colour of the bar shows whether the residue is in the well-defined core (black) or in the ill-defined residue ranges (cyan), as described in section 2 on ensemble composition.If well-defined core and ill-defined regions are not identified then it is shown as gray bars.

Random coil index (RCI) for chain A:



8 NMR restraints analysis

No restraints data found

9 Distance violation analysis (i)

No distance restraints data found

10 Dihedral-angle violation analysis (i)

Dihedral angle analysis failed due to data error in the dihedral angle restraints, possibly missing target value