



Full wwPDB NMR Structure Validation Report ⓘ

May 29, 2020 – 07:43 am BST

PDB ID : 5ML1
Title : NMR Structure of the Littorina littorea metallothionein, a snail MT folding into three distinct domains
Authors : Baumann, C.; Beil, A.; Jurt, S.; Zerbe, O.
Deposited on : 2016-12-06

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 ⓘ symbol.

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

Cyrange : Kirchner and Güntert (2011)
NmrClust : Kelley et al. (1996)
MolProbity : 4.02b-467
Percentile statistics : 20191225.v01 (using entries in the PDB archive December 25th 2019)
RCI : v_1n_11_5_13_A (Berjanski et al., 2005)
PANAV : Wang et al. (2010)
ShiftChecker : 2.11
Ideal geometry (proteins) : Engh & Huber (2001)
Ideal geometry (DNA, RNA) : Parkinson et al. (1996)
Validation Pipeline (wwPDB-VP) : 2.11

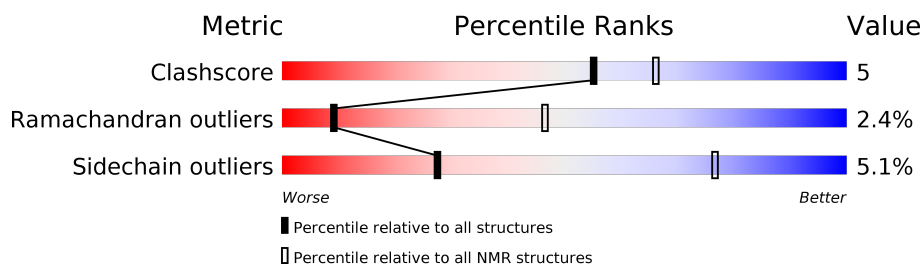
1 Overall quality at a glance

The following experimental techniques were used to determine the structure:

SOLUTION NMR

The overall completeness of chemical shifts assignment is 82%.

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 $\leq 5\%$

Mol	Chain	Length	Quality of chain
1	A	102	 86% 7% 7%

2 Ensemble composition and analysis

This entry contains 20 models. Model 7 is the overall representative, medoid model (most similar to other models).

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:6-A:38 (33)	0.29	4
2	A:41-A:102 (62)	0.52	7

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 3 clusters and 1 single-model cluster was found.

Cluster number	Models
1	2, 3, 4, 5, 6, 8, 9, 13, 14, 15, 16, 17, 20
2	10, 18, 19
3	1, 7, 12
Single-model clusters	11

3 Entry composition

There are 2 unique types of molecules in this entry. The entry contains 1289 atoms, of which 602 are hydrogens and 0 are deuteriums.

- Molecule 1 is a protein called Putative metallothionein.

Mol	Chain	Residues	Atoms						Trace
1	A	102	Total	C	H	N	O	S	0
			1280	380	602	124	146	28	

There are 2 discrepancies between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	Comment	Reference
A	1	GLY	-	expression tag	UNP Q962G0
A	2	SER	-	expression tag	UNP Q962G0

- Molecule 2 is CADMIUM ION (three-letter code: CD) (formula: Cd).

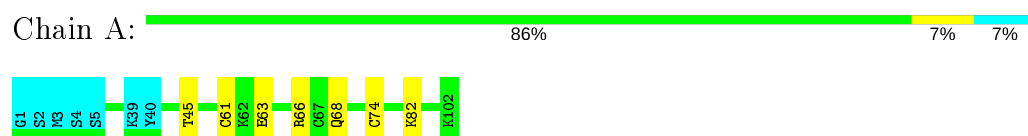
Mol	Chain	Residues	Atoms	
2	A	9	Total	Cd
			9	9

4 Residue-property plots [i](#)

4.1 Average score per residue in the NMR ensemble

These plots are provided for all protein, RNA and DNA 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: Putative metallothionein

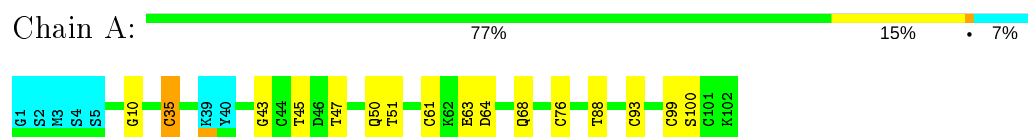


4.2 Scores per residue for each member of the ensemble

Colouring as in section [4.1](#) above.

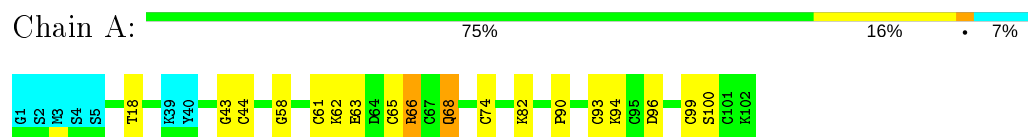
4.2.1 Score per residue for model 1

- Molecule 1: Putative metallothionein



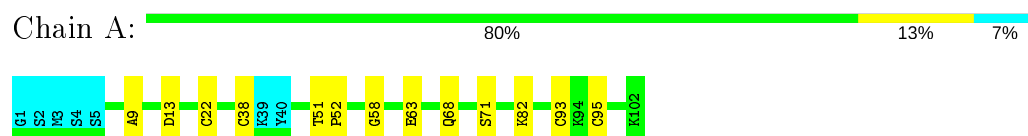
4.2.2 Score per residue for model 2

- Molecule 1: Putative metallothionein



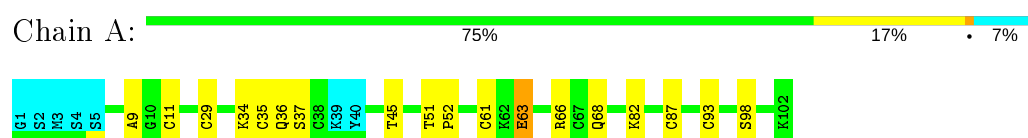
4.2.3 Score per residue for model 3

- Molecule 1: Putative metallothionein



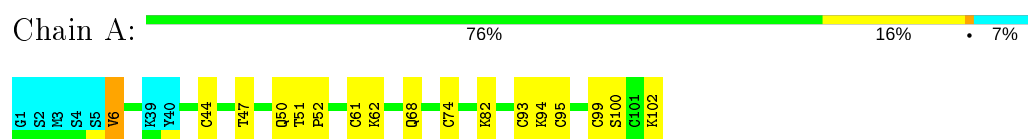
4.2.4 Score per residue for model 4

- Molecule 1: Putative metallothionein



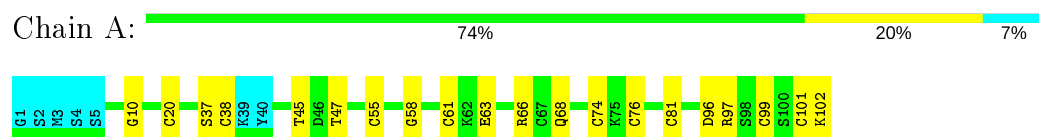
4.2.5 Score per residue for model 5

- Molecule 1: Putative metallothionein



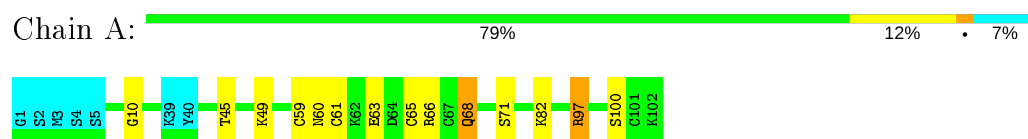
4.2.6 Score per residue for model 6

- Molecule 1: Putative metallothionein



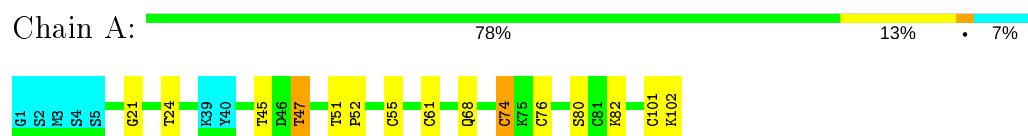
4.2.7 Score per residue for model 7 (medoid)

- Molecule 1: Putative metallothionein



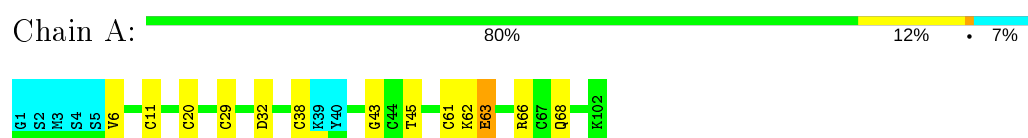
4.2.8 Score per residue for model 8

- Molecule 1: Putative metallothionein



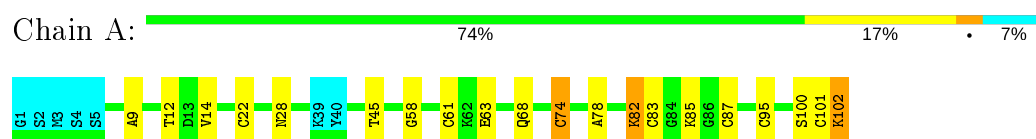
4.2.9 Score per residue for model 9

- Molecule 1: Putative metallothionein



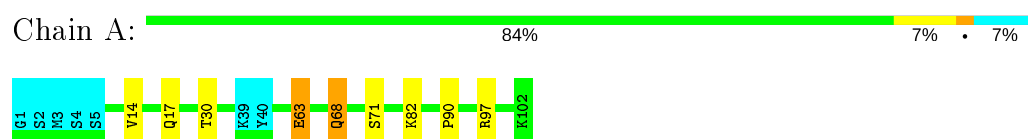
4.2.10 Score per residue for model 10

- Molecule 1: Putative metallothionein



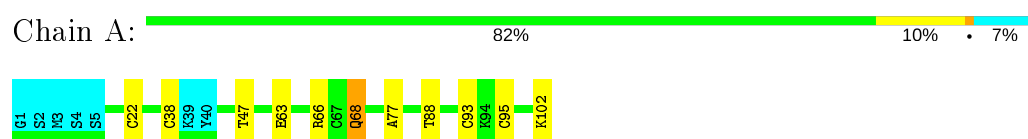
4.2.11 Score per residue for model 11

- Molecule 1: Putative metallothionein



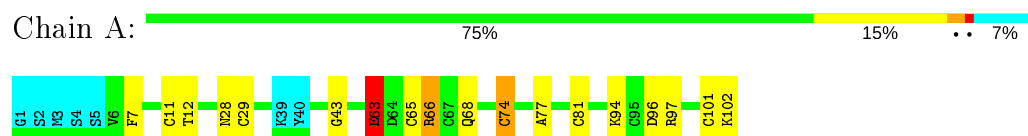
4.2.12 Score per residue for model 12

- Molecule 1: Putative metallothionein



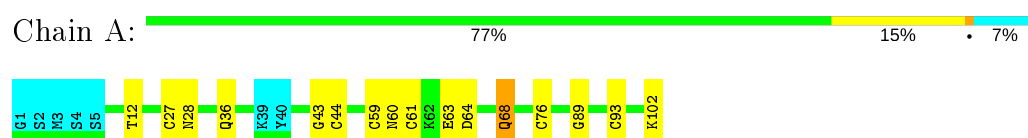
4.2.13 Score per residue for model 13

- Molecule 1: Putative metallothionein



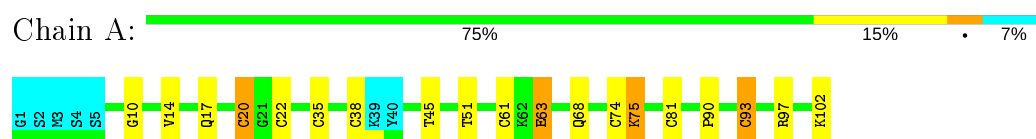
4.2.14 Score per residue for model 14

- Molecule 1: Putative metallothionein



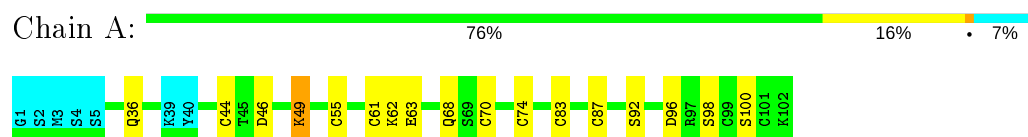
4.2.15 Score per residue for model 15

- Molecule 1: Putative metallothionein



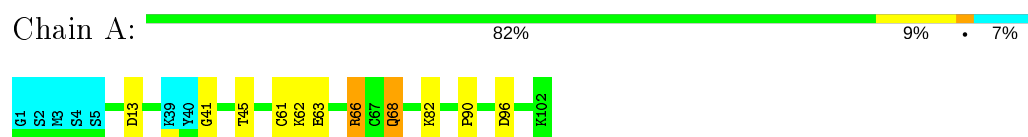
4.2.16 Score per residue for model 16

- Molecule 1: Putative metallothionein



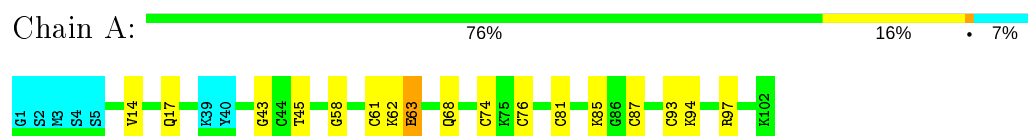
4.2.17 Score per residue for model 17

- Molecule 1: Putative metallothionein



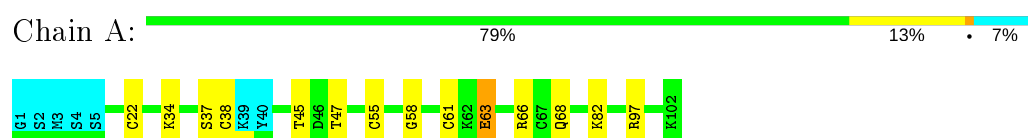
4.2.18 Score per residue for model 18

- Molecule 1: Putative metallothionein



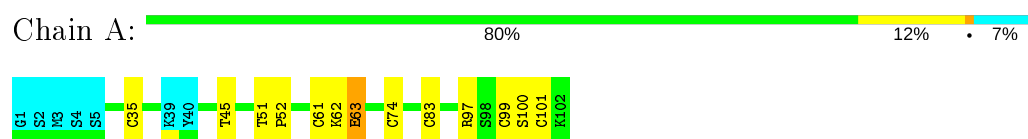
4.2.19 Score per residue for model 19

- Molecule 1: Putative metallothionein



4.2.20 Score per residue for model 20

- Molecule 1: Putative metallothionein



5 Refinement protocol and experimental data overview

Of the ? calculated structures, 20 were deposited, based on the following criterion: ?.

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

Software name	Classification	Version
CYANA	structure calculation	3.97
CNS-XPLOR	geometry optimization	

The following table shows chemical shift validation statistics as aggregates over all chemical shift files. Detailed validation can be found in section 6 of this report.

Chemical shift file(s)	input_cs.cif
Number of chemical shift lists	1
Total number of shifts	846
Number of shifts mapped to atoms	846
Number of unparsed shifts	0
Number of shifts with mapping errors	0
Number of shifts with mapping warnings	0
Assignment completeness (well-defined parts)	82%

No validations of the models with respect to experimental NMR restraints is performed at this time.

COVALENT-GEOMETRY INFOmissingINFO

5.1 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	627	554	554	6±2
All	All	12720	11080	11080	126

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 5.

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

Atom-1	Atom-2	Clash(Å)	Distance(Å)	Models	
				Worst	Total
1:A:74:CYS:SG	1:A:102:LYS:HE2	0.87	2.08	15	2
1:A:101:CYS:SG	1:A:102:LYS:N	0.74	2.60	13	2
1:A:14:VAL:HG23	1:A:17:GLN:NE2	0.60	2.11	15	3
1:A:68:GLN:HB2	1:A:90:PRO:O	0.59	1.98	2	3
1:A:68:GLN:HA	1:A:71:SER:OG	0.58	1.98	7	2
1:A:63:GLU:O	1:A:97:ARG:HB2	0.58	1.99	20	3
1:A:87:CYS:SG	1:A:93:CYS:HA	0.58	2.38	18	1
1:A:76:CYS:SG	1:A:101:CYS:HB2	0.58	2.37	6	1
1:A:74:CYS:SG	1:A:102:LYS:HE3	0.58	2.38	13	2
1:A:101:CYS:O	1:A:102:LYS:HB2	0.57	1.98	13	1
1:A:76:CYS:HB3	1:A:81:CYS:SG	0.57	2.40	18	1
1:A:74:CYS:SG	1:A:102:LYS:HB2	0.56	2.41	10	1
1:A:71:SER:HA	1:A:97:ARG:NH2	0.55	2.16	11	1
1:A:44:CYS:HA	1:A:61:CYS:SG	0.54	2.42	16	4
1:A:63:GLU:O	1:A:97:ARG:HG3	0.53	2.04	18	3
1:A:47:THR:HB	1:A:55:CYS:SG	0.53	2.43	8	3
1:A:85:LYS:HG3	1:A:95:CYS:SG	0.53	2.43	10	1
1:A:75:LYS:N	1:A:75:LYS:HD2	0.52	2.20	15	1
1:A:65:CYS:HB3	1:A:97:ARG:NH1	0.51	2.20	7	1
1:A:45:THR:HG22	1:A:61:CYS:SG	0.51	2.45	20	12
1:A:99:CYS:SG	1:A:101:CYS:HB3	0.51	2.46	20	1
1:A:7:PHE:HB3	1:A:11:CYS:SG	0.51	2.46	13	1
1:A:14:VAL:HG13	1:A:22:CYS:SG	0.51	2.46	10	2
1:A:12:THR:HG21	1:A:28:ASN:HB2	0.50	1.83	10	2
1:A:22:CYS:CB	1:A:38:CYS:HB3	0.50	2.36	19	1
1:A:66:ARG:HA	1:A:96:ASP:HA	0.49	1.84	13	4
1:A:76:CYS:HA	1:A:80:SER:O	0.49	2.08	8	1
1:A:11:CYS:HA	1:A:29:CYS:SG	0.48	2.47	13	2
1:A:22:CYS:HB2	1:A:38:CYS:CB	0.48	2.39	3	2
1:A:20:CYS:SG	1:A:38:CYS:HB3	0.47	2.48	6	3
1:A:96:ASP:OD2	1:A:98:SER:HB2	0.47	2.09	16	1
1:A:82:LYS:HE2	1:A:100:SER:O	0.47	2.08	7	1
1:A:83:CYS:HA	1:A:100:SER:HB2	0.47	1.87	10	1
1:A:35:CYS:SG	1:A:37:SER:HB2	0.47	2.50	4	1
1:A:97:ARG:CG	1:A:97:ARG:HH11	0.46	2.23	7	1
1:A:74:CYS:SG	1:A:102:LYS:HB3	0.46	2.50	8	1
1:A:83:CYS:HB3	1:A:101:CYS:CB	0.46	2.40	20	1
1:A:22:CYS:HB2	1:A:38:CYS:HB3	0.46	1.87	19	1
1:A:68:GLN:HB3	1:A:93:CYS:O	0.45	2.10	2	2
1:A:76:CYS:SG	1:A:81:CYS:HA	0.45	2.51	6	1
1:A:74:CYS:SG	1:A:102:LYS:HD3	0.45	2.51	5	1
1:A:68:GLN:NE2	1:A:68:GLN:H	0.45	2.09	3	1

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Atom-1	Atom-2	Clash(Å)	Distance(Å)	Models	
				Worst	Total
1:A:99:CYS:SG	1:A:100:SER:N	0.44	2.90	2	3
1:A:97:ARG:HH11	1:A:97:ARG:HG3	0.44	1.72	7	1
1:A:99:CYS:C	1:A:101:CYS:H	0.44	2.15	20	1
1:A:77:ALA:HA	1:A:88:THR:O	0.44	2.12	12	1
1:A:43:GLY:O	1:A:65:CYS:HB2	0.44	2.13	2	2
1:A:82:LYS:HG3	1:A:101:CYS:HA	0.43	1.90	8	1
1:A:43:GLY:HA2	1:A:62:LYS:HB2	0.43	1.90	2	3
1:A:83:CYS:HB2	1:A:101:CYS:SG	0.43	2.53	10	1
1:A:62:LYS:HE2	1:A:62:LYS:HA	0.43	1.91	16	2
1:A:59:CYS:SG	1:A:60:ASN:N	0.43	2.92	14	2
1:A:71:SER:HA	1:A:97:ARG:CZ	0.43	2.43	11	1
1:A:83:CYS:HB2	1:A:100:SER:HB2	0.43	1.90	16	1
1:A:37:SER:HA	1:A:66:ARG:NH2	0.42	2.29	19	1
1:A:87:CYS:O	1:A:92:SER:HB2	0.42	2.14	16	1
1:A:62:LYS:HA	1:A:62:LYS:HE2	0.42	1.92	5	1
1:A:12:THR:CG2	1:A:28:ASN:HB2	0.42	2.45	13	1
1:A:55:CYS:HB2	1:A:70:CYS:HB3	0.42	1.90	16	1
1:A:12:THR:HG21	1:A:28:ASN:CB	0.42	2.45	14	1
1:A:90:PRO:HA	1:A:93:CYS:HB2	0.42	1.92	15	1
1:A:6:VAL:HG21	1:A:66:ARG:NH2	0.41	2.30	9	1
1:A:51:THR:HA	1:A:52:PRO:C	0.41	2.35	3	1
1:A:21:GLY:HA2	1:A:24:THR:OG1	0.41	2.15	8	1
1:A:68:GLN:H	1:A:68:GLN:CD	0.41	2.18	14	1
1:A:55:CYS:HA	1:A:58:GLY:O	0.41	2.15	19	1
1:A:89:GLY:O	1:A:93:CYS:HB2	0.41	2.14	14	1
1:A:47:THR:O	1:A:50:GLN:HG3	0.41	2.15	5	2
1:A:82:LYS:H	1:A:82:LYS:HD2	0.41	1.76	10	1
1:A:43:GLY:HA3	1:A:64:ASP:HB3	0.41	1.90	1	1
1:A:97:ARG:HA	1:A:102:LYS:HG2	0.41	1.93	6	1
1:A:74:CYS:HB2	1:A:76:CYS:SG	0.41	2.56	18	1
1:A:85:LYS:HB3	1:A:87:CYS:SG	0.40	2.56	10	1
1:A:76:CYS:HB3	1:A:88:THR:HA	0.40	1.94	1	1
1:A:18:THR:HG22	1:A:94:LYS:HD3	0.40	1.93	2	1
1:A:11:CYS:HA	1:A:29:CYS:HB3	0.40	1.91	9	1
1:A:97:ARG:HA	1:A:102:LYS:O	0.40	2.17	13	1
1:A:87:CYS:SG	1:A:93:CYS:HB3	0.40	2.57	4	1
1:A:20:CYS:HB2	1:A:37:SER:HB3	0.40	1.93	6	1
1:A:46:ASP:HA	1:A:49:LYS:HG2	0.40	1.94	16	1

5.2 Torsion angles [i](#)

5.2.1 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	94/102 (92%)	78±3 (83±3%)	14±2 (15±3%)	2±1 (2±1%)	9	46
All	All	1880/2040 (92%)	1552 (83%)	282 (15%)	46 (2%)	9	46

All 13 unique Ramachandran outliers are listed below. They are sorted by the frequency of occurrence in the ensemble.

Mol	Chain	Res	Type	Models (Total)
1	A	63	GLU	17
1	A	51	THR	6
1	A	58	GLY	5
1	A	10	GLY	4
1	A	52	PRO	4
1	A	9	ALA	3
1	A	77	ALA	1
1	A	43	GLY	1
1	A	74	CYS	1
1	A	6	VAL	1
1	A	41	GLY	1
1	A	100	SER	1
1	A	78	ALA	1

5.2.2 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	76/82 (93%)	72±1 (95±2%)	4±1 (5±2%)	27	77
All	All	1520/1640 (93%)	1443 (95%)	77 (5%)	27	77

All 27 unique residues with a non-rotameric sidechain are listed below. They are sorted by the

frequency of occurrence in the ensemble.

Mol	Chain	Res	Type	Models (Total)
1	A	68	GLN	18
1	A	82	LYS	8
1	A	66	ARG	6
1	A	93	CYS	4
1	A	74	CYS	4
1	A	63	GLU	4
1	A	102	LYS	3
1	A	95	CYS	3
1	A	36	GLN	3
1	A	94	LYS	3
1	A	47	THR	2
1	A	49	LYS	2
1	A	13	ASP	2
1	A	81	CYS	2
1	A	35	CYS	1
1	A	30	THR	1
1	A	20	CYS	1
1	A	98	SER	1
1	A	76	CYS	1
1	A	27	CYS	1
1	A	64	ASP	1
1	A	99	CYS	1
1	A	62	LYS	1
1	A	85	LYS	1
1	A	32	ASP	1
1	A	75	LYS	1
1	A	34	LYS	1

5.2.3 RNA ⓘ

There are no RNA molecules in this entry.

5.3 Non-standard residues in protein, DNA, RNA chains ⓘ

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

5.4 Carbohydrates ⓘ

There are no carbohydrates in this entry.

5.5 Ligand geometry [i](#)

Of 9 ligands modelled in this entry, 9 are monoatomic - leaving 0 for Mogul analysis.

5.6 Other polymers [i](#)

There are no such molecules in this entry.

5.7 Polymer linkage issues [i](#)

There are no chain breaks in this entry.

6 Chemical shift validation [i](#)

The completeness of assignment taking into account all chemical shift lists is 82% for the well-defined parts and 78% for the entire structure.

6.1 Chemical shift list 1

File name: input_cs.cif

Chemical shift list name: *shiftFile_1*

6.1.1 Bookkeeping [i](#)

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	846
Number of shifts mapped to atoms	846
Number of unparsed shifts	0
Number of shifts with mapping errors	0
Number of shifts with mapping warnings	0
Number of shift outliers (ShiftChecker)	3

6.1.2 Chemical shift referencing [i](#)

The following table shows the suggested chemical shift referencing corrections.

Nucleus	# values	Correction \pm precision, ppm	Suggested action
$^{13}\text{C}_\alpha$	98	-3.53 ± 0.31	Should be applied
$^{13}\text{C}_\beta$	85	-2.43 ± 0.14	Should be applied
$^{13}\text{C}'$	0	—	None (insufficient data)
^{15}N	93	-0.83 ± 0.42	None needed (imprecise)

6.1.3 Completeness of resonance assignments [i](#)

The following table shows the completeness of the chemical shift assignments for the well-defined regions of the structure. The overall completeness is 82%, i.e. 749 atoms were assigned a chemical shift out of a possible 918. 2 out of 2 assigned methyl groups (LEU and VAL) were assigned stereospecifically.

	Total	^1H	^{13}C	^{15}N
Backbone	372/469 (79%)	186/187 (99%)	95/190 (50%)	91/92 (99%)
Sidechain	377/440 (86%)	244/266 (92%)	127/153 (83%)	6/21 (29%)

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	Total	¹ H	¹³ C	¹⁵ N
Aromatic	0/9 (0%)	0/5 (0%)	0/4 (0%)	0/0 (—%)
Overall	749/918 (82%)	430/458 (94%)	222/347 (64%)	97/113 (86%)

The following table shows the completeness of the chemical shift assignments for the full structure. The overall completeness is 78%, i.e. 777 atoms were assigned a chemical shift out of a possible 994. 2 out of 2 assigned methyl groups (LEU and VAL) were assigned stereospecifically.

	Total	¹ H	¹³ C	¹⁵ N
Backbone	382/504 (76%)	191/201 (95%)	98/204 (48%)	93/99 (94%)
Sidechain	395/473 (84%)	256/287 (89%)	133/164 (81%)	6/22 (27%)
Aromatic	0/17 (0%)	0/9 (0%)	0/8 (0%)	0/0 (—%)
Overall	777/994 (78%)	447/497 (90%)	231/376 (61%)	99/121 (82%)

6.1.4 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.

Mol	Chain	Res	Type	Atom	Shift, ppm	Expected range, ppm	Z-score
1	A	49	LYS	HG2	-0.48	2.67 – 0.07	-7.1
1	A	16	LYS	HG2	-0.41	2.67 – 0.07	-6.8
1	A	102	LYS	CE	46.32	46.00 – 37.80	5.4

6.1.5 Random Coil Index (RCI) plots ⓘ

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.

Random coil index (RCI) for chain A:

