wwPDB NMR Structure Validation Summary Report (i)

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PDB ID	:	5L6R
Title	:	PrP226* - Solution-state NMR structure of truncated human prion protein
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This is a wwPDB NMR Structure Validation Summary Report for a publicly released PDB entry.

We welcome your comments at *validation@mail.wwpdb.org*
A user guide is available at
http://wwpdb.org/validation/2016/NMRValidationReportHelp
with specific help available everywhere you see the (i) symbol.

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

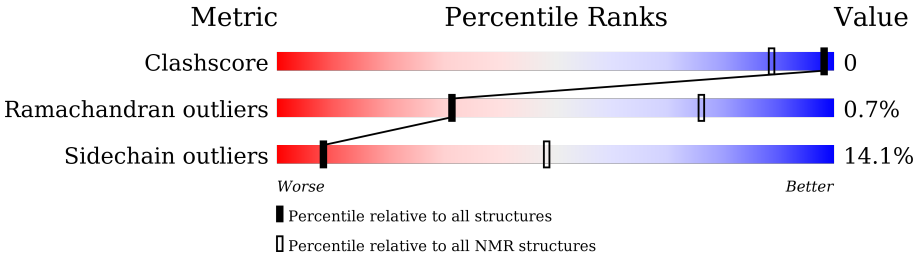
Cyrange	:	Kirchner and Güntert (2011)
NmrClust	:	Kelley et al. (1996)
MolProbity	:	4.02b-467
Mogul	:	unknown
Percentile statistics	:	20151230.v01 (using entries in the PDB archive December 30th 2015)
RCI	:	v_1n_11_5_13_A (Berjanski et al., 2005)
PANAV	:	Wang et al. (2010)
ShiftChecker	:	rb-20028442
Ideal geometry (proteins)	:	Engh & Huber (2001)
Ideal geometry (DNA, RNA)	:	Parkinson et al. (1996)
Validation Pipeline (wwPDB-VP)	:	rb-20028442

1 Overall quality at a glance (i)

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

The overall completeness of chemical shifts assignment is 78%.

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	114402	11133
Ramachandran outliers	111179	9975
Sidechain outliers	111093	9958

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	145	60% 12% • 28%

2 Ensemble composition and analysis (i)

This entry contains 20 models. Model 8 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:105-A:113 (9)	0.64	3
2	A:122-A:165, A:175-A:226 (96)	0.28	8

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 5 clusters. No single-model clusters were found.

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

3 Entry composition (i)

There is only 1 type of molecule in this entry. The entry contains 2249 atoms, of which 1087 are hydrogens and 0 are deuteriums.

- Molecule 1 is a protein called Major prion protein.

Mol	Chain	Residues	Atoms						Trace
			Total	C	H	N	O	S	
1	A	145	2249	716	1087	215	220	11	0

There are 8 discrepancies between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	Comment	Reference
A	227	GLY	-	expression tag	UNP P04156
A	228	GLY	-	expression tag	UNP P04156
A	229	HIS	-	expression tag	UNP P04156
A	230	HIS	-	expression tag	UNP P04156
A	231	HIS	-	expression tag	UNP P04156
A	232	HIS	-	expression tag	UNP P04156
A	233	HIS	-	expression tag	UNP P04156
A	234	HIS	-	expression tag	UNP P04156

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: Major prion protein

Chain A: 60% 12% • 28%

G90 Q91 G92 G93 G94 T95 H96 S97 Q98 W99 N100 K101 P102 S103 K104 T107 G114 A115 A116 A117 A118 G119 A120 V121 S132 R136 S143 R151 R156 V161 R164 P165 M166 D167 E168 Y169 S170 N171 Q172 N173 N174 F175 K185 T191 T192 K204 M205 M206

E207 R208 C214 Q217 Y218 E219 G227 G228 H229 H230 H231 H232 H233 H234

4.2 Residue scores for the representative (medoid) model from the NMR ensemble

The representative model is number 8. Colouring as in section 4.1 above.

- Molecule 1: Major prion protein

Chain A: 60% 11% • 28%

G90 Q91 G92 G93 G94 T95 H96 S97 Q98 W99 N100 K101 P102 S103 K104 T107 G114 A115 A116 A117 A118 G119 A120 V121 S132 R136 S143 R148 Y149 Y150 R151 V161 M166 D167 E168 Y169 S170 N171 Q172 N173 N174 C179 K185 Q186 T191 T192 M206 E207

R208 C214 Q217 Y218 E219 G227 G228 H229 H230 H231 H232 H233 H234

5 Refinement protocol and experimental data overview (i)

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
TALOS	geometry optimization	
YASARA	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)	5l6r_cs.cif
Number of chemical shift lists	1
Total number of shifts	1435
Number of shifts mapped to atoms	1435
Number of unparsed shifts	0
Number of shifts with mapping errors	0
Number of shifts with mapping warnings	0
Assignment completeness (well-defined parts)	78%

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

6 Model quality (i)

6.1 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 (average) 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	0.90±0.01	2±1/889 (0.3±0.1%)	0.95±0.02	4±2/1199 (0.3±0.1%)
All	All	0.90	48/17780 (0.3%)	0.95	83/23980 (0.3%)

All unique bond outliers are listed below. They are sorted according to the Z-score of the worst occurrence in the ensemble.

Mol	Chain	Res	Type	Atoms	Z	Observed(Å)	Ideal(Å)	Models	
								Worst	Total
1	A	161	VAL	CA-CB	7.66	1.70	1.54	2	20
1	A	161	VAL	CB-CG1	7.65	1.69	1.52	19	20
1	A	217	GLN	CB-CG	5.41	1.67	1.52	14	7
1	A	134	MET	CB-CG	-5.19	1.34	1.51	19	1

5 of 10 unique angle outliers are listed below. They are sorted according to the Z-score of the worst occurrence in the ensemble.

Mol	Chain	Res	Type	Atoms	Z	Observed(^{o})	Ideal(^{o})	Models	
								Worst	Total
1	A	164	ARG	NE-CZ-NH1	6.55	123.58	120.30	7	5
1	A	151	ARG	NE-CZ-NH1	6.54	123.57	120.30	3	12
1	A	136	ARG	NE-CZ-NH1	6.51	123.56	120.30	7	14
1	A	220	ARG	NE-CZ-NH1	6.45	123.52	120.30	1	5
1	A	156	ARG	NE-CZ-NH1	6.07	123.33	120.30	20	13

There are no chirality outliers.

There are no planarity outliers.

6.2 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 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	867	830	825	1±1
All	All	17340	16600	16500	17

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 1.

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

Atom-1	Atom-2	Clash(Å)	Distance(Å)	Models	
				Worst	Total
1:A:139:ILE:HG13	1:A:209:VAL:HG12	0.47	1.87	18	3
1:A:136:ARG:CZ	1:A:216:THR:HG21	0.46	2.41	14	1
1:A:132:SER:H	1:A:217:GLN:CD	0.43	2.18	6	10
1:A:175:PHE:CE2	1:A:214:CYS:SG	0.42	3.12	11	3

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	105/145 (72%)	97±1 (92±1%)	8±1 (7±1%)	1±1 (1±1%)	31	76
All	All	2100/2900 (72%)	1932 (92%)	154 (7%)	14 (1%)	31	76

All 4 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	176	VAL	9
1	A	165	PRO	3
1	A	175	PHE	1
1	A	196	GLU	1

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	95/122 (78%)	82±2 (86±2%)	13±2 (14±2%)	8 48
All	All	1900/2440 (78%)	1633 (86%)	267 (14%)	8 48

5 of 38 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	217	GLN	20
1	A	206	MET	20
1	A	204	LYS	19
1	A	132	SER	19
1	A	191	THR	18

6.3.3 RNA (i)

There are no RNA molecules in this entry.

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

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

6.5 Carbohydrates (i)

There are no carbohydrates in this entry.

6.6 Ligand geometry (i)

There are no ligands in this entry.

6.7 Other polymers (i)

There are no such molecules in this entry.

6.8 Polymer linkage issues (i)

There are no chain breaks in this entry.

7 Chemical shift validation (i)

The completeness of assignment taking into account all chemical shift lists is 78% for the well-defined parts and 71% for the entire structure.

7.1 Chemical shift list 1

File name: 5l6r_cs.cif

Chemical shift list name: *PrP226_chemical_shifts.str*

7.1.1 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	1435
Number of shifts mapped to atoms	1435
Number of unparsed shifts	0
Number of shifts with mapping errors	0
Number of shifts with mapping warnings	0
Number of shift outliers (ShiftChecker)	0

7.1.2 Chemical shift referencing (i)

The following table shows the suggested chemical shift referencing corrections.

Nucleus	# values	Correction ± precision, *ppm*	Suggested action
$^{13}C_{\alpha}$	127	-0.10 ± 0.18	None needed (< 0.5 ppm)
$^{13}C_{\beta}$	112	0.20 ± 0.09	None needed (< 0.5 ppm)
$^{13}C'$	0	—	None (insufficient data)
^{15}N	129	-0.33 ± 0.09	None needed (< 0.5 ppm)

7.1.3 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 78%, i.e. 1052 atoms were assigned a chemical shift out of a possible 1341. 10 out of 10 assigned methyl groups (LEU and VAL) were assigned stereospecifically.

	Total	^{1}H	^{13}C	^{15}N
Backbone	400/517 (77%)	200/206 (97%)	101/210 (48%)	99/101 (98%)
Sidechain	537/677 (79%)	336/401 (84%)	191/240 (80%)	10/36 (28%)

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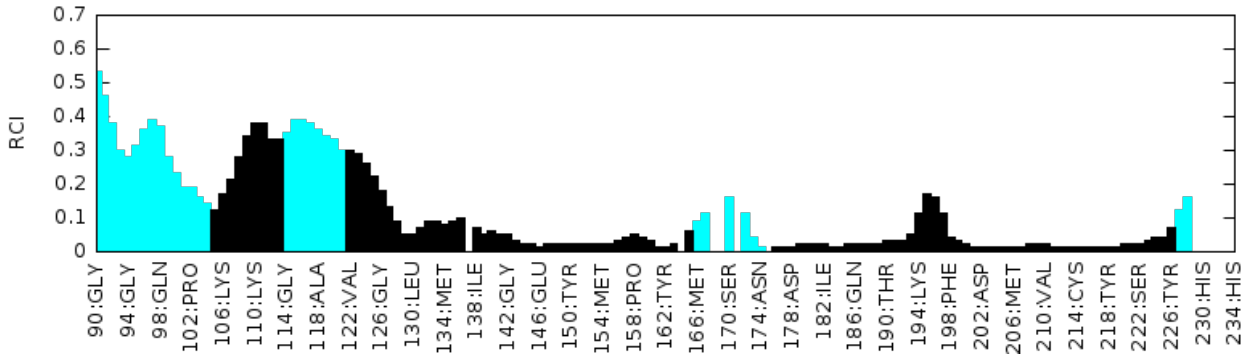
	Total	^{1}H	^{13}C	^{15}N
Aromatic	115/147 (78%)	59/75 (79%)	56/62 (90%)	0/10 (0%)
Overall	1052/1341 (78%)	595/682 (87%)	348/512 (68%)	109/147 (74%)

7.1.4 Statistically unusual chemical shifts (i)

There are no statistically unusual chemical shifts.

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.

Random coil index (RCI) for chain A: