Full wwPDB NMR Structure Validation Report (i)

May 29, 2020 – 12:03 am BST

PDB ID	:	2MRI
Title	:	Solution structure of a proteasome related subunit C terminal domain
Authors	:	Wu, Y.; Hu, Y.; Jin, C.
Deposited on	:	2014-07-06

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 (i) symbol.

The following versions of software and data (see references (1)) were used in the production of this report:

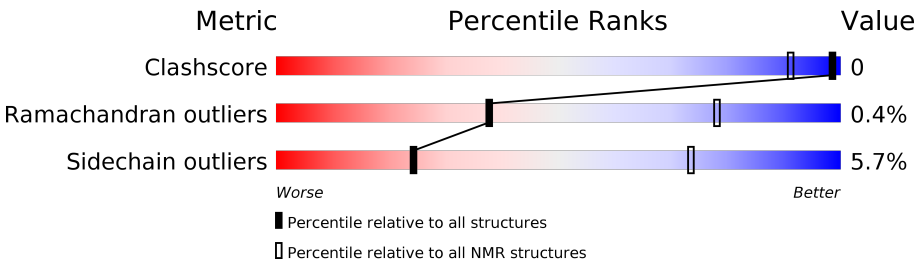
Cyrange	:	Kirchner and Güntert (2011)
NmrClust	:	Kelley et al. (1996)
MolProbity	:	4.02b-467
Percentile statistics	:	20191225.v01 (using entries in the PDB archive December 25th 2019)
RCI	:	v_1n_11_5_13_A (Berjanski et al., 2005)
PANAV	:	Wang et al. (2010)
ShiftChecker	:	2.11
Ideal geometry (proteins)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

1 Overall quality at a glance (i)

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

The overall completeness of chemical shifts assignment is 92%.

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	176	92% • 6%

2 Ensemble composition and analysis (i)

This entry contains 20 models. Model 13 is the overall representative, medoid model (most similar to other models). The authors have identified model 1 as representative, based on the following criterion: *lowest energy*.

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:184-A:197, A:205-A:355 (165)	0.63	13

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 4 clusters and 4 single-model clusters were found.

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

3 Entry composition (i)

There is only 1 type of molecule in this entry. The entry contains 2878 atoms, of which 1451 are hydrogens and 0 are deuteriums.

- Molecule 1 is a protein called 26S proteasome regulatory subunit RPN9.

Mol	Chain	Residues	Atoms						Trace
			Total	C	H	N	O	S	
1	A	176	2878	926	1451	230	266	5	0

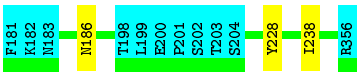
4 Residue-property plots (i)

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: 26S proteasome regulatory subunit RPN9

Chain A:



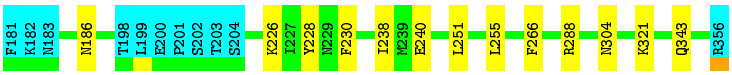
4.2 Scores per residue for each member of the ensemble

Colouring as in section 4.1 above.

4.2.1 Score per residue for model 1

- Molecule 1: 26S proteasome regulatory subunit RPN9

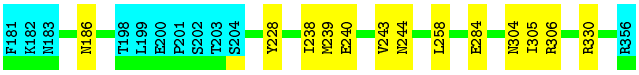
Chain A:



4.2.2 Score per residue for model 2

- Molecule 1: 26S proteasome regulatory subunit RPN9

Chain A:



4.2.3 Score per residue for model 3

- Molecule 1: 26S proteasome regulatory subunit RPN9

Chain A:

4.2.4 Score per residue for model 4

- Molecule 1: 26S proteasome regulatory subunit RPN9

Chain A:

4.2.5 Score per residue for model 5

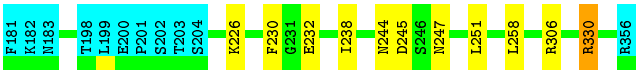
- Molecule 1: 26S proteasome regulatory subunit RPN9

Chain A:

4.2.6 Score per residue for model 6

- Molecule 1: 26S proteasome regulatory subunit RPN9

Chain A:



4.2.7 Score per residue for model 7

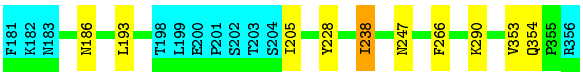
- Molecule 1: 26S proteasome regulatory subunit RPN9

Chain A:

4.2.8 Score per residue for model 8

- Molecule 1: 26S proteasome regulatory subunit RPN9

Chain A:



4.2.9 Score per residue for model 9

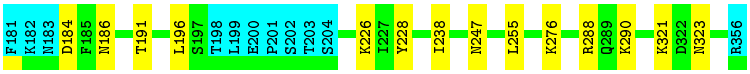
- Molecule 1: 26S proteasome regulatory subunit RPN9

Chain A:

4.2.10 Score per residue for model 10

- Molecule 1: 26S proteasome regulatory subunit RPN9

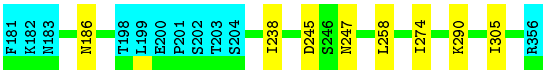
Chain A:



4.2.11 Score per residue for model 11

- Molecule 1: 26S proteasome regulatory subunit RPN9

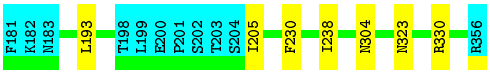
Chain A:



4.2.12 Score per residue for model 12

- Molecule 1: 26S proteasome regulatory subunit RPN9

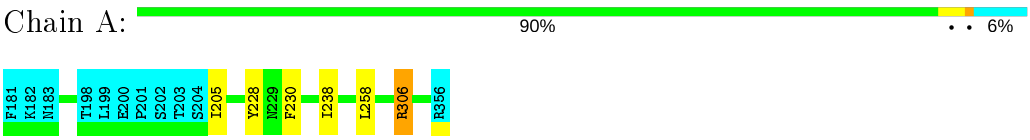
Chain A:



4.2.13 Score per residue for model 13 (medoid)

- Molecule 1: 26S proteasome regulatory subunit RPN9

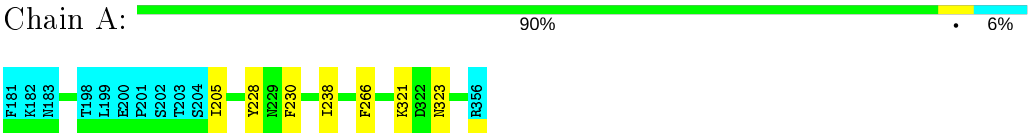
Chain A:



4.2.14 Score per residue for model 14

- Molecule 1: 26S proteasome regulatory subunit RPN9

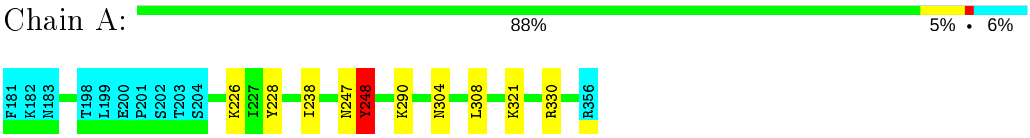
Chain A:



4.2.15 Score per residue for model 15

- Molecule 1: 26S proteasome regulatory subunit RPN9

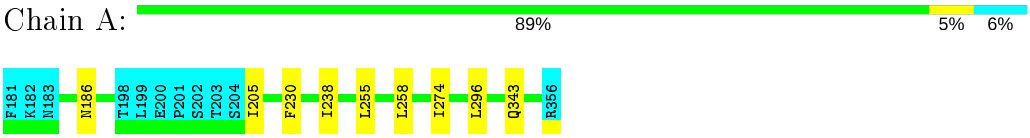
Chain A:



4.2.16 Score per residue for model 16

- Molecule 1: 26S proteasome regulatory subunit RPN9

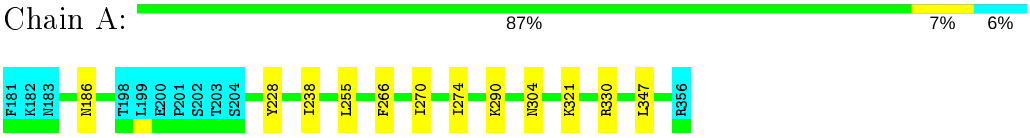
Chain A:



4.2.17 Score per residue for model 17

- Molecule 1: 26S proteasome regulatory subunit RPN9

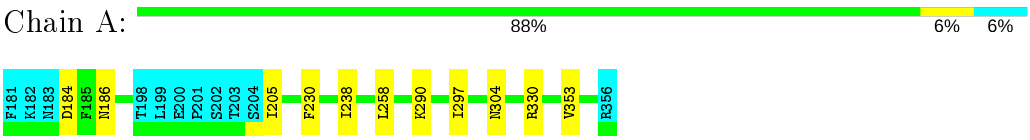
Chain A:



4.2.18 Score per residue for model 18

- Molecule 1: 26S proteasome regulatory subunit RPN9

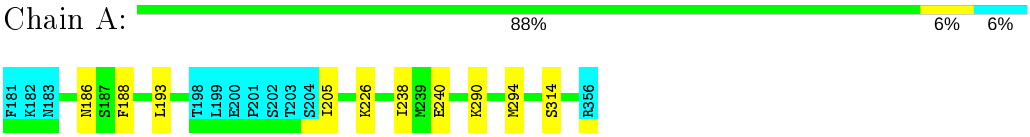
Chain A:



4.2.19 Score per residue for model 19

- Molecule 1: 26S proteasome regulatory subunit RPN9

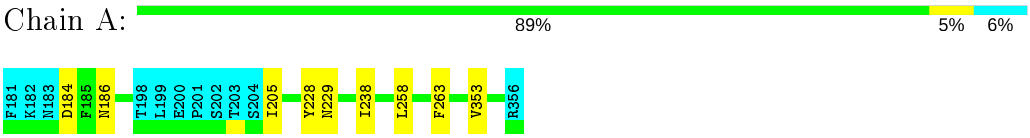
Chain A:



4.2.20 Score per residue for model 20

- Molecule 1: 26S proteasome regulatory subunit RPN9

Chain A:



5 Refinement protocol and experimental data overview (i)

The models were refined using the following method: *simulated annealing*.

Of the 100 calculated structures, 20 were deposited, based on the following criterion: *structures with the lowest energy*.

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

Software name	Classification	Version
AMBER	refinement	

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	2216
Number of shifts mapped to atoms	2216
Number of unparsed shifts	0
Number of shifts with mapping errors	0
Number of shifts with mapping warnings	0
Assignment completeness (well-defined parts)	92%

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

COVALENT-GEOMETRY INFOmissingINFO

5.1 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	1337	1360	1360	0±1
All	All	26740	27200	27200	7

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 0.

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

Atom-1	Atom-2	Clash(Å)	Distance(Å)	Models	
				Worst	Total
1:A:218:SER:CB	1:A:251:LEU:HD11	0.51	2.36	7	1
1:A:239:MET:O	1:A:243:VAL:HG13	0.45	2.12	3	1
1:A:297:ILE:HG23	1:A:353:VAL:HG21	0.44	1.89	18	1
1:A:239:MET:O	1:A:243:VAL:HG23	0.42	2.14	2	1
1:A:243:VAL:HG12	1:A:252:PHE:CD1	0.42	2.49	3	1
1:A:227:ILE:H	1:A:227:ILE:HD12	0.42	1.74	7	1
1:A:248:TYR:CD1	1:A:248:TYR:C	0.41	2.93	15	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	165/176 (94%)	158±2 (95±1%)	7±2 (4±1%)	1±1 (0±0%)	38	78
All	All	3300/3520 (94%)	3150 (95%)	137 (4%)	13 (0%)	38	78

All 5 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	230	PHE	8
1	A	205	ILE	2
1	A	238	ILE	1
1	A	226	LYS	1
1	A	228	TYR	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	153/164 (93%)	144±2 (94±2%)	9±2 (6±2%)	24	73

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Mol	Chain	Analysed	Rotameric	Outliers	Percentiles
All	All	3060/3280 (93%)	2887 (94%)	173 (6%)	24 73

All 47 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	238	ILE	20
1	A	186	ASN	13
1	A	290	LYS	9
1	A	258	LEU	9
1	A	205	ILE	8
1	A	330	ARG	7
1	A	304	ASN	7
1	A	321	LYS	7
1	A	266	PHE	6
1	A	247	ASN	6
1	A	251	LEU	5
1	A	228	TYR	5
1	A	184	ASP	5
1	A	226	LYS	4
1	A	305	ILE	4
1	A	255	LEU	4
1	A	240	GLU	4
1	A	343	GLN	3
1	A	306	ARG	3
1	A	193	LEU	3
1	A	323	ASN	3
1	A	274	ILE	3
1	A	196	LEU	3
1	A	308	LEU	2
1	A	244	ASN	2
1	A	314	SER	2
1	A	245	ASP	2
1	A	263	PHE	2
1	A	232	GLU	2
1	A	288	ARG	2
1	A	353	VAL	2
1	A	284	GLU	1
1	A	297	ILE	1
1	A	229	ASN	1
1	A	188	PHE	1
1	A	276	LYS	1

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Mol	Chain	Res	Type	Models (Total)
1	A	325	GLU	1
1	A	354	GLN	1
1	A	286	PHE	1
1	A	294	MET	1
1	A	236	HIS	1
1	A	223	LEU	1
1	A	248	TYR	1
1	A	347	LEU	1
1	A	225	ASP	1
1	A	270	ILE	1
1	A	296	LEU	1

5.2.3 RNA (i)

There are no RNA molecules in this entry.

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

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

5.4 Carbohydrates (i)

There are no carbohydrates in this entry.

5.5 Ligand geometry (i)

There are no ligands in this entry.

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 92% for the well-defined parts and 91% for the entire structure.

6.1 Chemical shift list 1

File name: input_cs.cif

Chemical shift list name: *assigned_chem_shift_list_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	2216
Number of shifts mapped to atoms	2216
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 ± precision, *ppm*	Suggested action
$^{13}C_{\alpha}$	175	-0.67 ± 0.14	Should be applied
$^{13}C_{\beta}$	170	0.14 ± 0.10	None needed (< 0.5 ppm)
$^{13}C'$	171	-0.28 ± 0.18	None needed (< 0.5 ppm)
^{15}N	166	0.42 ± 0.27	None needed (< 0.5 ppm)

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 92%, i.e. 1925 atoms were assigned a chemical shift out of a possible 2093. 0 out of 36 assigned methyl groups (LEU and VAL) were assigned stereospecifically.

	Total	^{1}H	^{13}C	^{15}N
Backbone	811/817 (99%)	324/326 (99%)	328/330 (99%)	159/161 (99%)
Sidechain	988/1088 (91%)	598/631 (95%)	372/420 (89%)	18/37 (49%)

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	Total	^{1}H	^{13}C	^{15}N
Aromatic	126/188 (67%)	64/101 (63%)	60/80 (75%)	2/7 (29%)
Overall	1925/2093 (92%)	986/1058 (93%)	760/830 (92%)	179/205 (87%)

The following table shows the completeness of the chemical shift assignments for the full structure. The overall completeness is 91%, i.e. 2031 atoms were assigned a chemical shift out of a possible 2235. 0 out of 37 assigned methyl groups (LEU and VAL) were assigned stereospecifically.

	Total	^{1}H	^{13}C	^{15}N
Backbone	853/870 (98%)	341/347 (98%)	346/352 (98%)	166/171 (97%)
Sidechain	1048/1168 (90%)	636/679 (94%)	393/447 (88%)	19/42 (45%)
Aromatic	130/197 (66%)	66/106 (62%)	62/84 (74%)	2/7 (29%)
Overall	2031/2235 (91%)	1043/1132 (92%)	801/883 (91%)	187/220 (85%)

6.1.4 Statistically unusual chemical shifts (i)

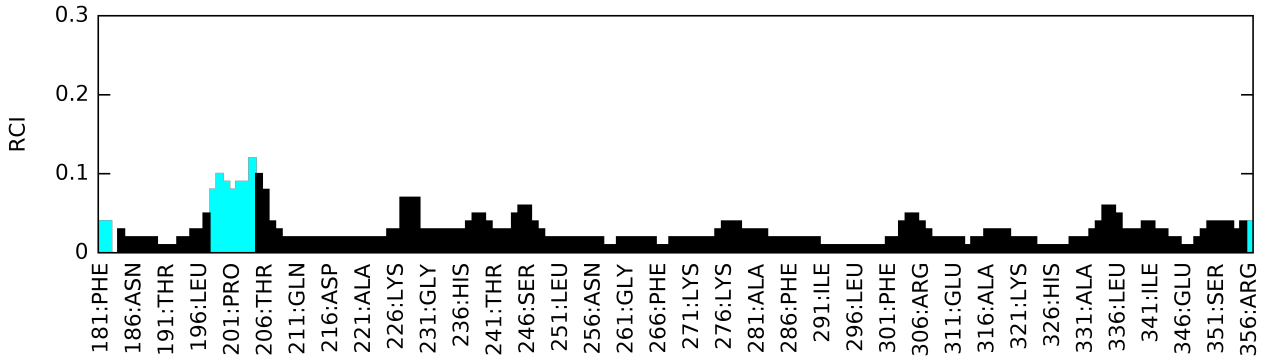
The following table lists the statistically unusual chemical shifts. These are statistical measures, and large deviations from the mean do not necessarily imply incorrect assignments. Molecules containing paramagnetic centres or hemes are expected to give rise to anomalous chemical shifts.

Mol	Chain	Res	Type	Atom	Shift, *ppm*	Expected range, *ppm*	Z-score
1	A	346	GLU	HA	2.01	6.30 – 2.20	-5.5
1	A	238	ILE	H	11.87	11.73 – 4.83	5.2
1	A	251	LEU	HB2	-0.14	3.32 – -0.08	-5.2

6.1.5 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:


RCI
0.3
0.2
0.1
0
181:PHE
186:ASN
191:THR
196:LEU
201:PRO
206:THR
211:GLN
216:ASP
221:ALA
226:LYS
231:GLY
236:HIS
241:THR
246:SER
251:LEU
256:ASN
261:GLY
266:PHE
271:LYS
276:LYS
281:ALA
286:PHE
291:ILE
296:LEU
301:PHE
306:ARG
311:GLU
316:ALA
321:LYS
326:HIS
331:ALA
336:LEU
341:ILE
346:GLU
351:SER
356:ARG