wwPDB NMR Structure Validation Summary Report

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PDB ID	:	2LWE
BMRB ID	:	18623
Title	:	Solution structure of mutant (T170E) second CARD of human RIG-I
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Deposited on	:	2012-07-27

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
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:

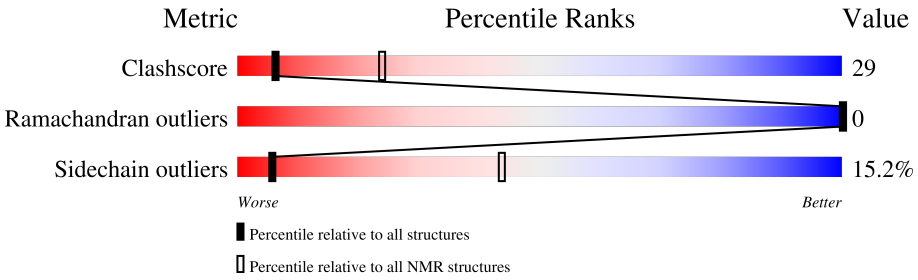
MolProbity	:	4.02b-467
Percentile statistics	:	20191225.v01 (using entries in the PDB archive December 25th 2019)
wwPDB-RCI	:	v_1n_11_5_13_A (Berjanski et al., 2005)
PANAV	:	Wang et al. (2010)
wwPDB-ShiftChecker	:	v1.2
Ideal geometry (proteins)	:	Engh & Huber (2001)
Ideal geometry (DNA, RNA)	:	Parkinson et al. (1996)
Validation Pipeline (wwPDB-VP)	:	2.36.2

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 85%.

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	100	40% 44% 5% 7% .

2 Ensemble composition and analysis

This entry contains 20 models. Model 3 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:100-A:188 (89)	0.40	3

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 and 1 single-model cluster was found.

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

3 Entry composition (i)

There is only 1 type of molecule in this entry. The entry contains 1472 atoms, of which 677 are hydrogens and 0 are deuteriums.

- Molecule 1 is a protein called Probable ATP-dependent RNA helicase DDX58.

Mol	Chain	Residues	Atoms						Trace
			Total	C	H	N	O	S	
1	A	96	1472	507	677	132	150	6	0

There are 5 discrepancies between the modelled and reference sequences:

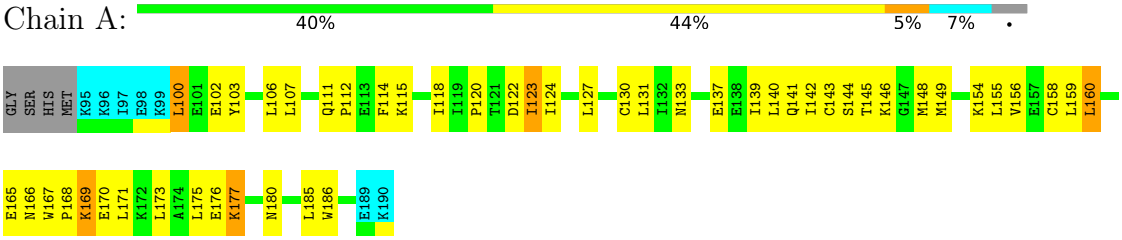
Chain	Residue	Modelled	Actual	Comment	Reference
A	91	GLY	-	expression tag	UNP O95786
A	92	SER	-	expression tag	UNP O95786
A	93	HIS	-	expression tag	UNP O95786
A	94	MET	-	expression tag	UNP O95786
A	170	GLU	THR	engineered mutation	UNP O95786

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: Probable ATP-dependent RNA helicase DDX58



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

The representative model is number 3. Colouring as in section 4.1 above.

• Molecule 1: Probable ATP-dependent RNA helicase DDX58

5 Refinement protocol and experimental data overview

The models were refined using the following method: *DGSA-distance geometry simulated annealing.*

Of the 2100 calculated structures, 20 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
CNS	structure solution	
CNS	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	1206
Number of shifts mapped to atoms	1088
Number of unparsed shifts	0
Number of shifts with mapping errors	118
Number of shifts with mapping warnings	0
Assignment completeness (well-defined parts)	85%

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	732	623	764	43±5
All	All	14640	12460	15280	859

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 29.

5 of 246 unique clashes are listed below, sorted by their clash magnitude.

Atom-1	Atom-2	Clash(Å)	Distance(Å)	Models	
				Worst	Total
1:A:185:LEU:H	1:A:185:LEU:HD13	0.87	1.29	1	4
1:A:142:ILE:HG23	1:A:146:LYS:HB2	0.85	1.47	5	20
1:A:106:LEU:HG	1:A:186:TRP:CZ3	0.78	2.14	6	4
1:A:124:ILE:HG21	1:A:140:LEU:HD21	0.75	1.58	20	3
1:A:169:LYS:O	1:A:173:LEU:HB2	0.74	1.83	15	9

6.3 Torsion angles

6.3.1 Protein backbone

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	89/100 (89%)	83±1 (93±1%)	6±1 (7±1%)	0±0 (0±0%)	100	100
All	All	1780/2000 (89%)	1655 (93%)	125 (7%)	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	84/94 (89%)	71±3 (85±4%)	13±3 (15±4%)	6	44
All	All	1680/1880 (89%)	1425 (85%)	255 (15%)	6	44

5 of 42 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	123	ILE	20
1	A	177	LYS	19
1	A	160	LEU	18
1	A	176	GLU	16
1	A	169	LYS	15

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 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 85% for the well-defined parts and 84% 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	1206
Number of shifts mapped to atoms	1088
Number of unparsed shifts	0
Number of shifts with mapping errors	118
Number of shifts with mapping warnings	0
Number of shift outliers (ShiftChecker)	2

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

- No matching atom found in the structure. First 5 (of 118) occurrences are reported below.

List ID	Chain	Res	Type	Atom	Shift Data		
					Value	Uncertainty	Ambiguity
1	A	93	HIS	HA	4.596	0.03	1
1	A	93	HIS	HB2	3.108	0.04	2
1	A	93	HIS	HB3	3.177	0.04	2
1	A	93	HIS	HD2	7.1885	0.04	1
1	A	93	HIS	HE1	8.2922	0.04	1
1	A	93	HIS	C	175.2566	0.09	1
1	A	93	HIS	CA	56.5347	0.40	1
1	A	93	HIS	CB	30.1	0.40	1
1	A	93	HIS	CD2	119.862	0.35	1
1	A	93	HIS	CE1	137.1764	0.35	1
1	A	94	MET	H	8.3047	0.01	1
1	A	94	MET	HA	4.353	0.03	1
1	A	94	MET	HB2	1.965	0.04	2
1	A	94	MET	HG2	2.49	0.04	2

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List ID	Chain	Res	Type	Atom	Shift Data		
					Value	Uncertainty	Ambiguity
1	A	94	MET	HG3	2.44	0.04	2
1	A	94	MET	C	176.2397	0.09	1
1	A	94	MET	CA	55.9042	0.40	1
1	A	94	MET	CB	32.9316	0.40	1
1	A	94	MET	CG	31.875	0.35	1
1	A	94	MET	N	121.595	0.05	1
1	A	95	LYS	HD2	1.675	0.04	2
1	A	96	LYS	HG2	1.35	0.04	2
1	A	97	ILE	HG12	1.24	0.04	2
1	A	100	LEU	HB2	1.73	0.04	2
1	A	101	GLU	HB2	2.033	0.04	2
1	A	102	GLU	HB2	1.66	0.04	2
1	A	102	GLU	HG2	1.88	0.04	2
1	A	103	TYR	HB2	2.572	0.04	2
1	A	104	ARG	HB2	1.986	0.04	2
1	A	104	ARG	HD2	3.42	0.04	2
1	A	105	LEU	HB2	1.511	0.04	2
1	A	106	LEU	HB2	1.56	0.04	2
1	A	107	LEU	HB2	2.18	0.04	2
1	A	108	LYS	HG2	1.63	0.04	2
1	A	109	ARG	HB2	2.315	0.04	2
1	A	109	ARG	HG2	1.99	0.04	2
1	A	109	ARG	HD2	3.412	0.04	2
1	A	110	LEU	HB2	2.23	0.04	2
1	A	111	GLN	HB2	2.36	0.04	2
1	A	112	PRO	HB2	1.893	0.04	2
1	A	112	PRO	HG2	2.007	0.04	2
1	A	112	PRO	HD2	3.86	0.04	2
1	A	113	GLU	HB2	2.02	0.04	2
1	A	113	GLU	HG2	2.363	0.04	2
1	A	114	PHE	HB2	2.418	0.04	2
1	A	115	LYS	HB2	1.75	0.04	2
1	A	117	ARG	HB2	1.541	0.04	2
1	A	117	ARG	HG2	1.549	0.04	2
1	A	118	ILE	HG12	-0.32	0.04	2
1	A	119	ILE	HG12	1.2	0.04	2
1	A	120	PRO	HB2	2.03	0.04	2
1	A	120	PRO	HG2	1.54	0.04	2
1	A	120	PRO	HD2	4.23	0.04	2
1	A	123	ILE	HG12	1.39	0.04	2
1	A	124	ILE	HG12	1.24	0.04	2

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List ID	Chain	Res	Type	Atom	Shift Data		
					Value	Uncertainty	Ambiguity
1	A	126	ASP	HB2	2.633	0.04	2
1	A	127	LEU	HB2	1.67	0.04	2
1	A	129	GLU	HB2	1.861	0.04	2
1	A	129	GLU	HG2	2.192	0.04	2
1	A	130	CYS	HB2	2.795	0.04	2
1	A	131	LEU	HB2	1.407	0.04	2
1	A	133	ASN	HB2	2.75	0.04	2
1	A	135	GLU	HB2	2.007	0.04	2
1	A	135	GLU	HG2	2.32	0.04	2
1	A	136	CYS	HB2	2.662	0.04	2
1	A	137	GLU	HG2	2.166	0.04	2
1	A	138	GLU	HB2	1.984	0.04	2
1	A	139	ILE	HG12	1.07	0.04	2
1	A	140	LEU	HB2	1.435	0.04	2
1	A	141	GLN	HG2	2.44	0.04	2
1	A	143	CYS	HB2	2.815	0.04	2
1	A	144	SER	HB2	3.96	0.04	2
1	A	146	LYS	HB2	1.712	0.04	2
1	A	146	LYS	HG2	1.52	0.04	2
1	A	146	LYS	HD2	1.545	0.04	2
1	A	148	MET	HB2	1.989	0.04	2
1	A	148	MET	HG2	2.84	0.04	2
1	A	149	MET	HB2	1.888	0.04	2
1	A	149	MET	HG2	2.53	0.04	2
1	A	153	GLU	HG2	2.076	0.04	2
1	A	154	LYS	HG2	1.206	0.04	2
1	A	154	LYS	HD2	1.57	0.04	2
1	A	154	LYS	HE2	2.72	0.04	2
1	A	155	LEU	HB2	1.284	0.04	2
1	A	157	GLU	HG2	2.029	0.04	2
1	A	158	CYS	HB2	2.632	0.04	2
1	A	159	LEU	HB2	-0.636	0.04	2
1	A	160	LEU	HB2	1.315	0.04	2
1	A	161	ARG	HB2	1.82	0.04	2
1	A	162	SER	HB2	3.68	0.04	2
1	A	163	ASP	HB2	2.71	0.04	2
1	A	164	LYS	HB2	2.17	0.04	2
1	A	164	LYS	HG2	1.645	0.04	2
1	A	166	ASN	HB2	2.833	0.04	2
1	A	167	TRP	HB2	3.25	0.04	2
1	A	168	PRO	HB2	0.876	0.04	2

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List ID	Chain	Res	Type	Atom	Shift Data		
					Value	Uncertainty	Ambiguity
1	A	168	PRO	HD2	2.158	0.04	2
1	A	169	LYS	HB2	1.85	0.04	2
1	A	169	LYS	HG2	1.335	0.04	2
1	A	170	GLU	HB2	1.969	0.04	2
1	A	170	GLU	HG2	2.128	0.04	2
1	A	171	LEU	HB2	1.474	0.04	2
1	A	172	LYS	HB2	2.008	0.04	2
1	A	173	LEU	HB2	1.532	0.04	2
1	A	175	LEU	HB2	1.403	0.04	2
1	A	176	GLU	HB2	2.052	0.04	2
1	A	176	GLU	HG2	2.273	0.04	2
1	A	177	LYS	HG2	1.552	0.04	2
1	A	177	LYS	HE2	2.92	0.04	2
1	A	178	GLU	HB2	1.806	0.04	2
1	A	178	GLU	HG2	2.49	0.04	2
1	A	179	ARG	HB2	2.008	0.04	2
1	A	180	ASN	HB2	2.563	0.04	2
1	A	182	PHE	HB2	2.685	0.04	2
1	A	185	LEU	HB2	1.06	0.04	2
1	A	186	TRP	HB2	2.672	0.04	2
1	A	187	ILE	HG12	0.97	0.04	2
1	A	189	GLU	HB2	1.806	0.04	2

7.1.2 Chemical shift referencing

The following table shows the suggested chemical shift referencing corrections.

Nucleus	# values	Correction ± precision, *ppm*	Suggested action
$^{13}C_{\alpha}$	98	-0.68 ± 0.13	Should be checked
$^{13}C_{\beta}$	95	0.32 ± 0.10	None needed (< 0.5 ppm)
$^{13}C'$	94	-0.52 ± 0.13	Should be applied
^{15}N	94	0.57 ± 0.32	None needed (imprecise)

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 85%, i.e. 1106 atoms were assigned a chemical shift out of a possible 1300. 0 out of 17 assigned methyl groups (LEU and VAL) were assigned stereospecifically.

	Total	^{1}H	^{13}C	^{15}N
Backbone	437/441 (99%)	176/177 (99%)	175/178 (98%)	86/86 (100%)
Sidechain	638/806 (79%)	434/521 (83%)	204/255 (80%)	0/30 (0%)
Aromatic	31/53 (58%)	16/26 (62%)	13/25 (52%)	2/2 (100%)
Overall	1106/1300 (85%)	626/724 (86%)	392/458 (86%)	88/118 (75%)

7.1.4 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.

List Id	Chain	Res	Type	Atom	Shift, *ppm*	Expected range, *ppm*	Z-score
1	A	168	PRO	HG3	-0.60	0.33 – 3.48	-8.0
1	A	159	LEU	HB2	-0.64	-0.07 – 3.30	-6.7

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: