# wwPDB NMR Structure Validation Summary Report (i)

Apr 20, 2024 – 12:04 PM EDT

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|---|---|---|
| PDB ID | : | 2MRE |
| BMRB ID | : | 25070 |
| Title | : | NMR structure of the Rad18-UBZ/ubiquitin complex |
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| Deposited on | : | 2014-07-03 |

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:

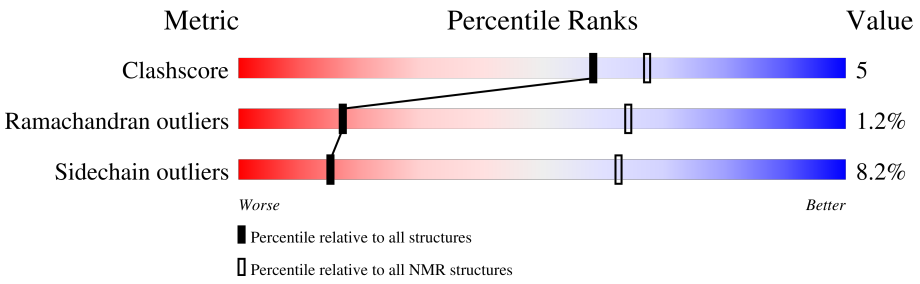
| | | |
|---|---|---|
| MolProbity | : | 4.02b-467 |
| Percentile statistics | : | 20191225.v01 (using entries in the PDB archive December 25th 2019) |
| wwPDB-RCI | : | v_1n_11_5_13_A (Berjanski et al., 2005) |
| PANAV | : | Wang et al. (2010) |
| wwPDB-ShiftChecker | : | v1.2 |
| Ideal geometry (proteins) | : | 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 90%.

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 | 79 | 76% 16% 8% |
| 2 | B | 33 | 58% 12% • 27% |

# 2 Ensemble composition and analysis (i)

This entry contains 20 models. Model 16 is the overall representative, medoid model (most similar to other models). The authors have identified model 1 as representative, based on the following criterion: *closest to the average*.

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:1-A:73, B:201-B:224 (97) | 0.41 | 16 |

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

# 3 Entry composition (i)

There are 3 unique types of molecules in this entry. The entry contains 1765 atoms, of which 892 are hydrogens and 0 are deuteriums.

- Molecule 1 is a protein called Polyubiquitin-C.

| Mol | Chain | Residues | Atoms | | | | | | Trace |
|---|---|---|---|---|---|---|---|---|---|
| | | | Total | C | H | N | O | S | |
| 1 | A | 79 | 1267 | 389 | 646 | 110 | 121 | 1 | 0 |

There are 3 discrepancies between the modelled and reference sequences:

| Chain | Residue | Modelled | Actual | Comment | Reference |
|---|---|---|---|---|---|
| A | -2 | GLY | - | expression tag | UNP P0CG48 |
| A | -1 | SER | - | expression tag | UNP P0CG48 |
| A | 0 | HIS | - | expression tag | UNP P0CG48 |

- Molecule 2 is a protein called E3 ubiquitin-protein ligase RAD18.

| Mol | Chain | Residues | Atoms | | | | | | Trace |
|---|---|---|---|---|---|---|---|---|---|
| | | | Total | C | H | N | O | S | |
| 2 | B | 33 | 497 | 150 | 246 | 48 | 50 | 3 | 0 |

There are 3 discrepancies between the modelled and reference sequences:

| Chain | Residue | Modelled | Actual | Comment | Reference |
|---|---|---|---|---|---|
| B | 195 | GLY | - | expression tag | UNP Q9NS91 |
| B | 196 | SER | - | expression tag | UNP Q9NS91 |
| B | 197 | ARG | - | expression tag | UNP Q9NS91 |

- Molecule 3 is ZINC ION (three-letter code: ZN) (formula: Zn).

| Mol | Chain | Residues | Atoms | |
|---|---|---|---|---|
| | | | Total | Zn |
| 3 | B | 1 | 1 | 1 |

# 4 Residue-property plots (i)

## 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: Polyubiquitin-C

Chain A: 76% 16% 8%

G-2 S-1 H0 M1 Q2 I3 T7 K11 L15 E16 V17 D21 V26 Q41 F45 L67 H68 L69 V70 R74 G75 G76

• Molecule 2: E3 ubiquitin-protein ligase RAD18

Chain B: 58% 12% • 27%

G195 S196 R197 Q198 V199 T200 C204 E213 I216 H219 L220 S225 R226 E227

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

The representative model is number 16. Colouring as in section 4.1 above.

• Molecule 1: Polyubiquitin-C

Chain A: 75% 16% • 8%

G-2 S-1 H0 M1 Q2 I3 T7 K11 L15 E16 V17 D21 V26 P38 Q41 L67 H68 L69 V70 L71 R72 L73 R74 G75 G76

• Molecule 2: E3 ubiquitin-protein ligase RAD18

Chain B: 58% 12% • 27%

G195 S196 R197 Q198 V199 T200 P205 E213 I216 H219 L220 S225 R226 E227

# 5 Refinement protocol and experimental data overview (i)

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

Of the 200 calculated structures, 20 were deposited, based on the following criterion: *target function*.

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

| Software name | Classification | Version |
|---|---|---|
| CYANA | structure solution | 2.1 |
| 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 | 1276 |
| Number of shifts mapped to atoms | 1276 |
| Number of unparsed shifts | 0 |
| Number of shifts with mapping errors | 0 |
| Number of shifts with mapping warnings | 0 |
| Assignment completeness (well-defined parts) | 90% |

# 6 Model quality (i)

## 6.1 Standard geometry (i)

Bond lengths and bond angles in the following residue types are not validated in this section: ZN

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 (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 | 582 | 609 | 608 | 6±2 |
| 2 | B | 180 | 177 | 176 | 3±1 |
| All | All | 15260 | 15720 | 15680 | 152 |

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.

5 of 37 unique clashes are listed below, sorted by their clash magnitude.

| Atom-1 | Atom-2 | Clash(Å) | Distance(Å) | Models | |
|---|---|---|---|---|---|
| | | | | Worst | Total |
| 1:A:42:ARG:HB3 | 1:A:70:VAL:HB | 0.60 | 1.73 | 11 | 5 |
| 1:A:17:VAL:HG21 | 1:A:26:VAL:HG22 | 0.59 | 1.75 | 14 | 20 |
| 1:A:7:THR:HG22 | 1:A:69:LEU:HD12 | 0.53 | 1.78 | 1 | 15 |
| 1:A:5:VAL:HG13 | 1:A:69:LEU:HB2 | 0.53 | 1.80 | 2 | 2 |
| 1:A:43:LEU:HG | 1:A:50:LEU:HD12 | 0.52 | 1.81 | 6 | 2 |

## 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 | 73/79 (92%) | 69±1 (94±1%) | 3±1 (4±1%) | 1±1 (2±1%) | 13 | 57 |
| 2 | B | 24/33 (73%) | 22±1 (92±4%) | 2±1 (8±4%) | 0±0 (0±0%) | 100 | 100 |
| All | All | 1940/2240 (87%) | 1823 (94%) | 94 (5%) | 23 (1%) | 17 | 64 |

All 3 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 | 21 | ASP | 16 |
| 1 | A | 22 | THR | 6 |
| 1 | A | 73 | LEU | 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 | 67/70 (96%) | 62±1 (93±1%) | 5±1 (7±1%) | 19 | 67 |
| 2 | B | 23/31 (74%) | 20±1 (88±3%) | 3±1 (12±3%) | 9 | 52 |
| All | All | 1800/2020 (89%) | 1653 (92%) | 147 (8%) | 15 | 62 |

5 of 20 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 | 3 | ILE | 20 |
| 1 | A | 67 | LEU | 20 |
| 2 | B | 216 | ILE | 20 |
| 2 | B | 219 | HIS | 20 |

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| Mol | Chain | Res | Type | Models (Total) |
|---|---|---|---|---|
| 1 | A | 41 | GLN | 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 (i)

Of 1 ligands modelled in this entry, 1 is monoatomic - leaving 0 for Mogul analysis.

## 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 90% 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 (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 | 1276 |
| Number of shifts mapped to atoms | 1276 |
| 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 |
|---|---|---|---|
| $^{13}C_{\alpha}$ | 103 | -0.47 ± 0.09 | None needed (< 0.5 ppm) |
| $^{13}C_{\beta}$ | 97 | 0.12 ± 0.12 | None needed (< 0.5 ppm) |
| $^{13}C'$ | 95 | -0.21 ± 0.16 | None needed (< 0.5 ppm) |
| $^{15}N$ | 98 | 0.27 ± 0.38 | 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 90%, i.e. 1204 atoms were assigned a chemical shift out of a possible 1345. 0 out of 18 assigned methyl groups (LEU and VAL) were assigned stereospecifically.

| | Total | $^{1}H$ | $^{13}C$ | $^{15}N$ |
|---|---|---|---|---|
| Backbone | 468/480 (98%) | 192/194 (99%) | 186/194 (96%) | 90/92 (98%) |
| Sidechain | 736/813 (91%) | 500/527 (95%) | 229/258 (89%) | 7/28 (25%) |

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| | Total | $^{1}$H | $^{13}$C | $^{15}$N |
|---|---|---|---|---|
| Aromatic | 0/52 (0%) | 0/26 (0%) | 0/21 (0%) | 0/5 (0%) |
| Overall | 1204/1345 (90%) | 692/747 (93%) | 415/473 (88%) | 97/125 (78%) |

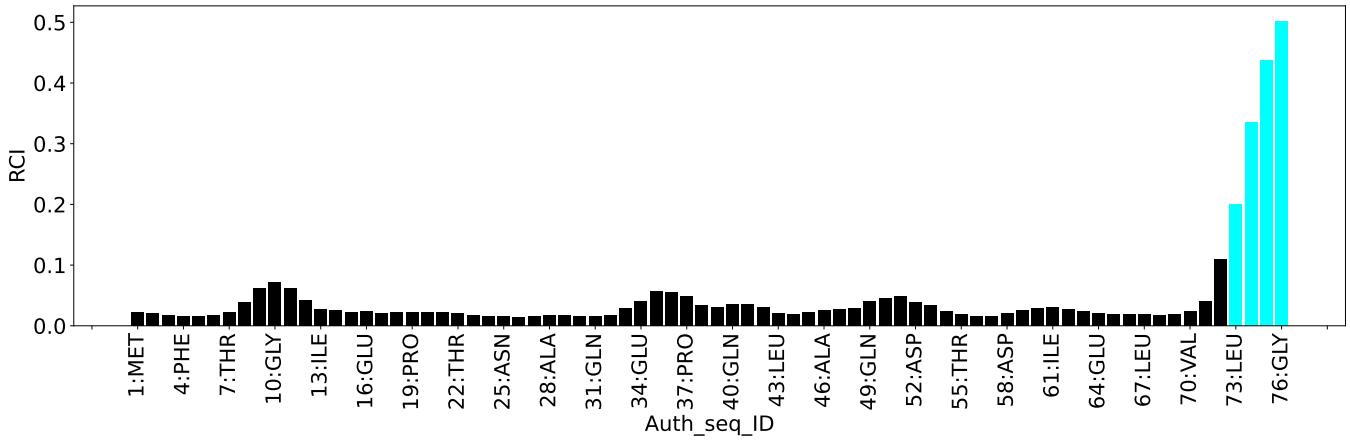
### 7.1.4 Statistically unusual chemical shifts (i)

There are no statistically unusual chemical shifts.

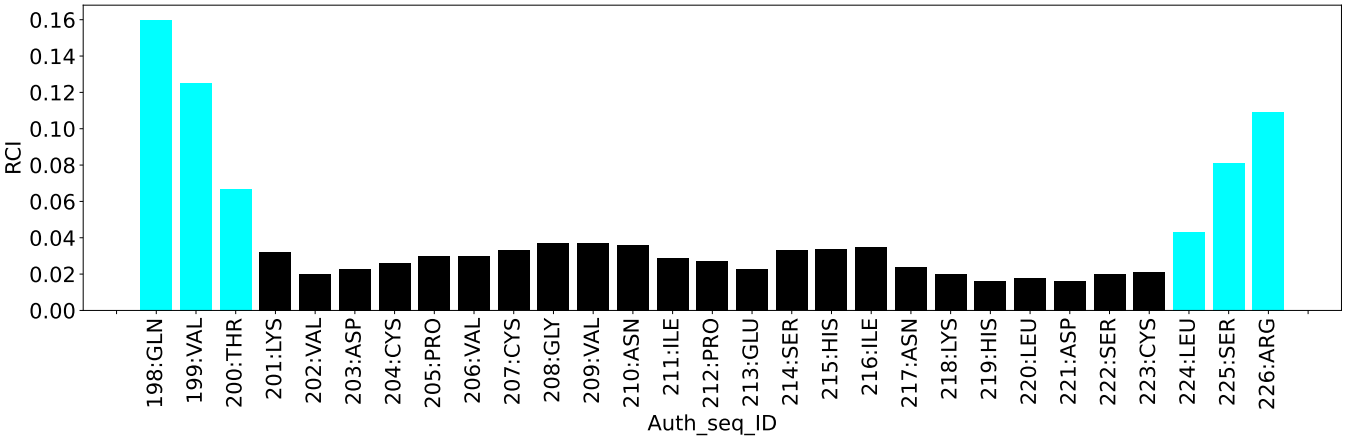
### 7.1.5 Random Coil Index (RCI) plots (i)

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.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:




Random coil index (RCI) for chain B:


RCI
0.16
0.14
0.12
0.10
0.08
0.06
0.04
0.02
0.00
198:GLN
199:VAL
200:THR
201:LYS
202:VAL
203:ASP
204:CYS
205:PRO
206:VAL
207:CYS
208:GLY
209:VAL
210:ASN
211:ILE
212:PRO
213:GLU
214:SER
215:HIS
216:ILE
217:ASN
218:LYS
219:HIS
220:LEU
221:ASP
222:SER
223:CYS
224:LEU
225:SER
226:ARG
Auth_seq_ID