



# Full wwPDB NMR Structure Validation Report ⓘ

Apr 26, 2016 – 08:43 PM BST

PDB ID : 2I85  
Title : NMR solution structure of Human ephrinB2 ectodomain  
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Deposited on : 2006-09-01

This is a Full wwPDB NMR Structure Validation Report for a publicly released PDB entry.  
We welcome your comments at [validation@mail.wwpdb.org](mailto:validation@mail.wwpdb.org)  
A user guide is available at  
<http://wwpdb.org/validation/2016/NMRValidationReportHelp>  
with specific help available everywhere you see the ⓘ symbol.

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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  
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-20027457  
Ideal geometry (proteins) : Engh & Huber (2001)  
Ideal geometry (DNA, RNA) : Parkinson et al. (1996)  
Validation Pipeline (wwPDB-VP) : rb-20027457

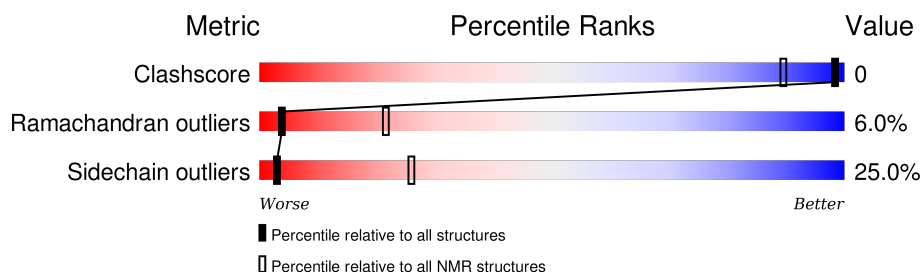
# 1 Overall quality at a glance

The following experimental techniques were used to determine the structure:

*SOLUTION NMR*

The overall completeness of chemical shifts assignment is 75%.

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  $\geq 3$ , 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	142	

## 2 Ensemble composition and analysis

This entry contains 10 models. Model 4 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:3-A:45, A:50-A:116, A:123-A:142 (130)	1.23	4

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 2 clusters and 2 single-model clusters were found.

Cluster number	Models
1	1, 2, 3, 4
2	5, 7, 9, 10
Single-model clusters	6; 8

### 3 Entry composition [i](#)

There is only 1 type of molecule in this entry. The entry contains 2060 atoms, of which 924 are hydrogens and 0 are deuteriums.

- Molecule 1 is a protein called Ephrin-B2.

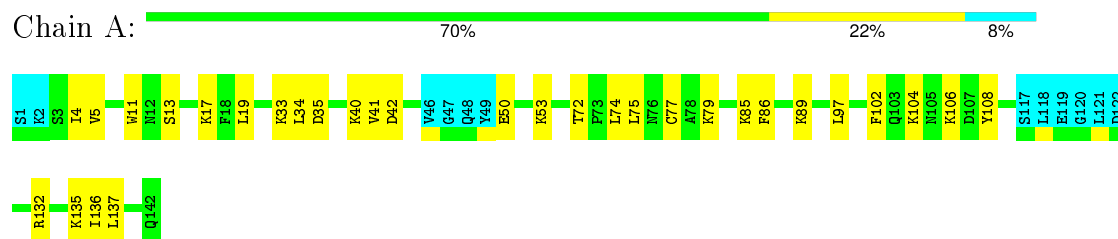
Mol	Chain	Residues	Atoms							Trace
1	A	142	Total	C	H	N	O	S		0
			2060	727	924	184	218	7		

## 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: Ephrin-B2

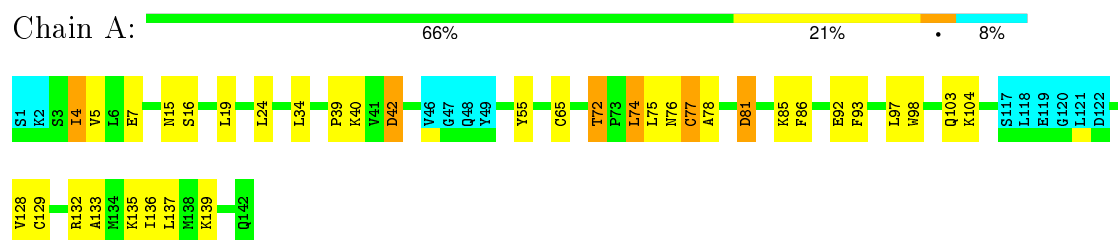


### 4.2 Scores per residue for each member of the ensemble

Colouring as in section 4.1 above.

#### 4.2.1 Score per residue for model 1

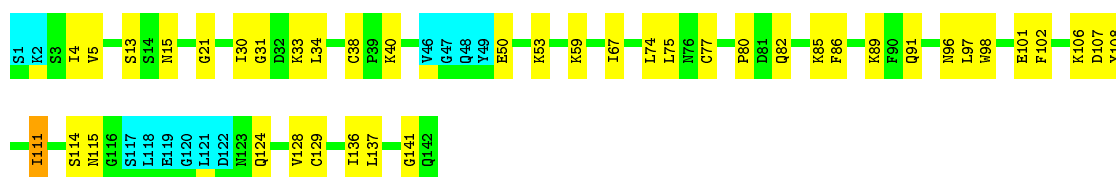
- Molecule 1: Ephrin-B2



#### 4.2.2 Score per residue for model 2

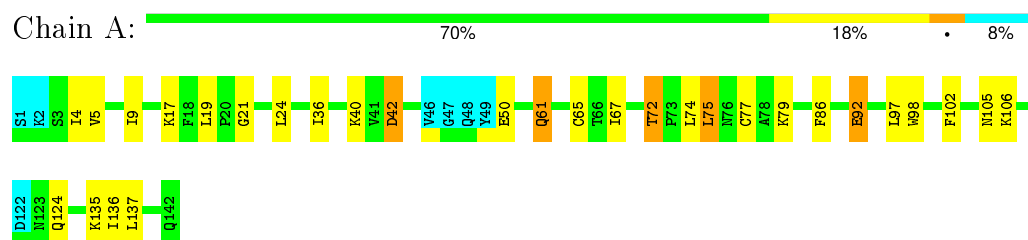
- Molecule 1: Ephrin-B2





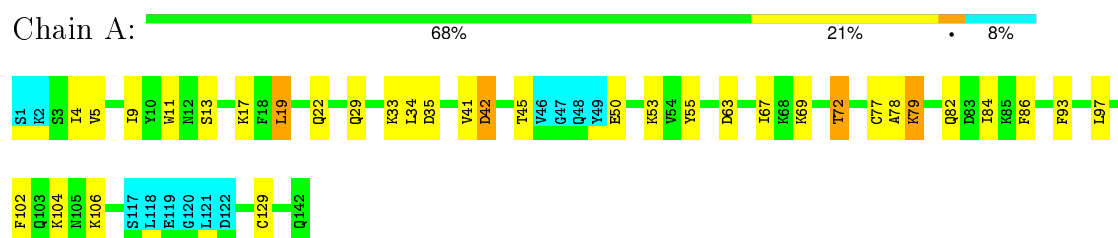
#### 4.2.3 Score per residue for model 3

- Molecule 1: Ephrin-B2



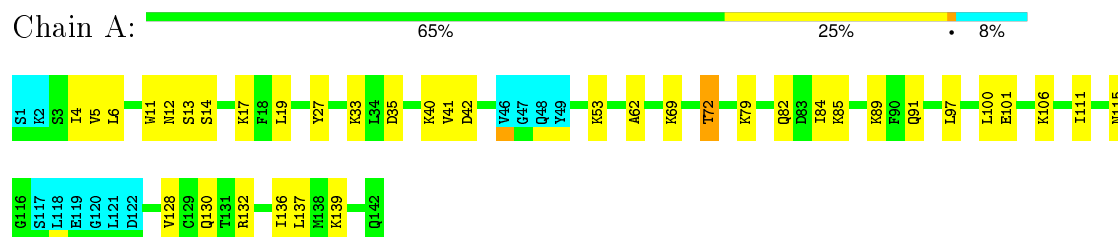
#### 4.2.4 Score per residue for model 4 (medoid)

- Molecule 1: Ephrin-B2



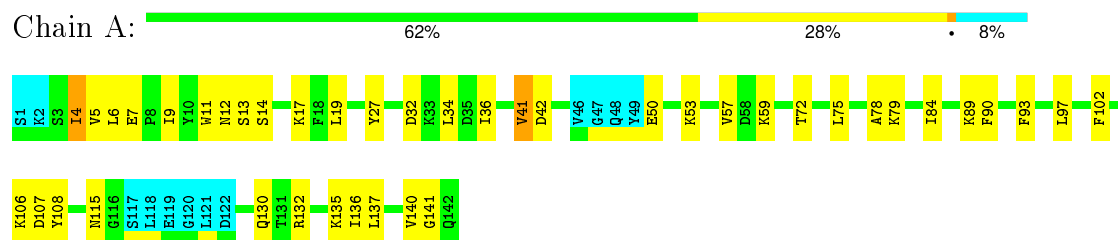
#### 4.2.5 Score per residue for model 5

- Molecule 1: Ephrin-B2



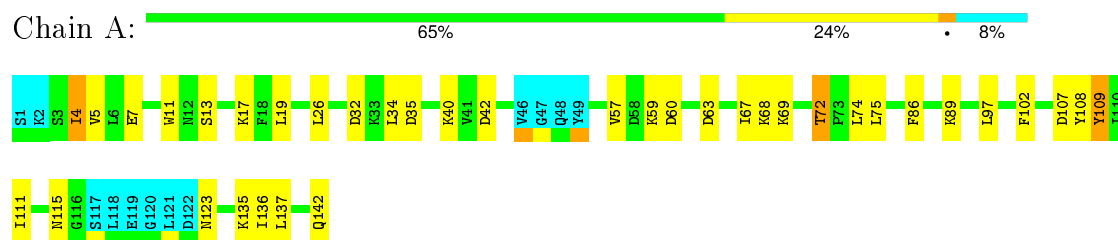
#### 4.2.6 Score per residue for model 6

- Molecule 1: Ephrin-B2



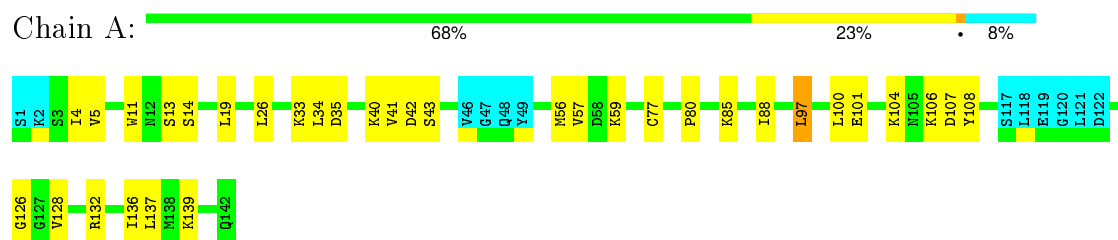
#### 4.2.7 Score per residue for model 7

- Molecule 1: Ephrin-B2



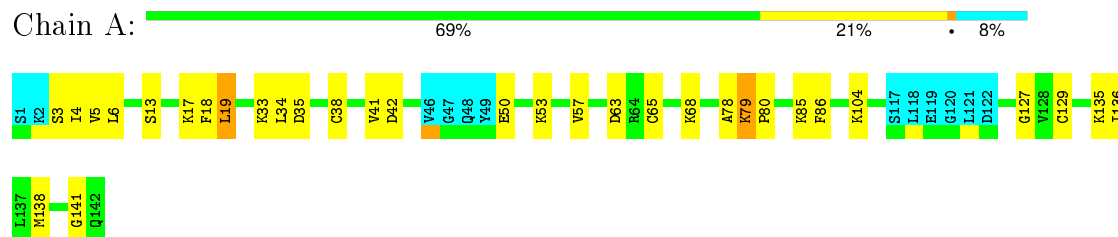
#### 4.2.8 Score per residue for model 8

- Molecule 1: Ephrin-B2



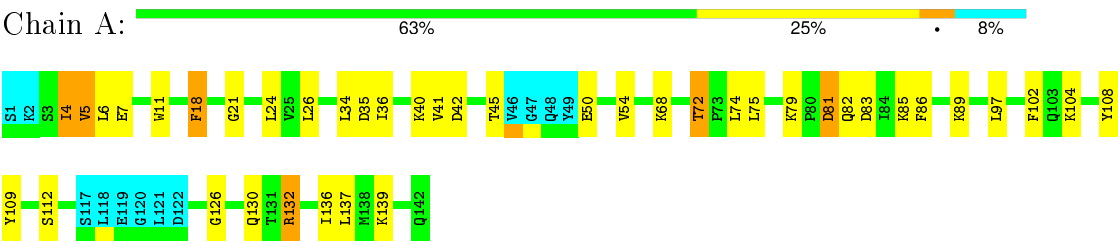
#### 4.2.9 Score per residue for model 9

- Molecule 1: Ephrin-B2



4.2.10 Score per residue for model 10

● Molecule 1: Ephrin-B2





## 5 Refinement protocol and experimental data overview

The models were refined using the following method: *distance geometry*.

Of the 200 calculated structures, 10 were deposited, based on the following criterion: *target funtion less than 2*.

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

Software name	Classification	Version
Cyana	structure solution	
Amber	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)	BMRB entry 7220
Number of chemical shift lists	1
Total number of shifts	1515
Number of shifts mapped to atoms	1439
Number of unparsed shifts	0
Number of shifts with mapping errors	76
Number of shifts with mapping warnings	0
Assignment completeness (well-defined parts)	75%

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](#)

There are no covalent bond-length or bond-angle outliers.

Chiral center outliers are detected by calculating the chiral volume of a chiral center and verifying if the center is modelled as a planar moiety or with the opposite hand. A planarity outlier is detected by checking planarity of atoms in a peptide group, atoms in a mainchain group or atoms of a sidechain that are expected to be planar.

Mol	Chain	Chirality	Planarity
1	A	0.0±0.0	0.2±0.4
All	All	0	2

There are no bond-length outliers.

There are no bond-angle outliers.

There are no chirality outliers.

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

Mol	Chain	Res	Type	Group	Models (Total)
1	A	108	TYR	Sidechain	1
1	A	55	TYR	Sidechain	1

### 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	1046	853	1048	1±1
All	All	10460	8530	10480	9

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:74:LEU:HD11	1:A:78:ALA:HB2	0.60	1.73	1	1
1:A:55:TYR:CD2	1:A:67:ILE:HG23	0.57	2.34	4	1
1:A:109:TYR:CD2	1:A:137:LEU:HD21	0.46	2.46	7	1
1:A:61:GLN:NE2	1:A:67:ILE:HD13	0.46	2.25	3	1
1:A:111:ILE:HD12	1:A:129:CYS:SG	0.46	2.50	2	1
1:A:67:ILE:HD13	1:A:111:ILE:HG23	0.45	1.88	7	1
1:A:18:PHE:CG	1:A:18:PHE:O	0.41	2.74	10	1
1:A:36:ILE:HD13	1:A:75:LEU:HD13	0.41	1.93	3	1
1:A:62:ALA:HA	1:A:111:ILE:HD12	0.40	1.93	5	1

## 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	129/142 (91%)	99±2 (77±1%)	22±3 (17±2%)	8±2 (6±2%)	4	21
All	All	1290/1420 (91%)	988 (77%)	224 (17%)	78 (6%)	4	21

All 32 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	5	VAL	9
1	A	42	ASP	8
1	A	72	THR	6
1	A	41	VAL	4
1	A	77	CYS	4
1	A	79	LYS	4
1	A	4	ILE	4
1	A	78	ALA	3
1	A	21	GLY	3
1	A	141	GLY	3
1	A	80	PRO	3
1	A	12	ASN	2
1	A	18	PHE	2
1	A	81	ASP	2

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Mol	Chain	Res	Type	Models (Total)
1	A	126	GLY	2
1	A	19	LEU	2
1	A	124	GLN	2
1	A	22	GLN	1
1	A	30	ILE	1
1	A	133	ALA	1
1	A	50	GLU	1
1	A	31	GLY	1
1	A	65	CYS	1
1	A	127	GLY	1
1	A	90	PHE	1
1	A	39	PRO	1
1	A	14	SER	1
1	A	11	TRP	1
1	A	97	LEU	1
1	A	13	SER	1
1	A	132	ARG	1
1	A	92	GLU	1

### 6.3.2 Protein sidechains ⓘ

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	119/129 (92%)	89±4 (75±3%)	30±4 (25±3%)	3	26
All	All	1190/1290 (92%)	892 (75%)	298 (25%)	3	26

All 87 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	4	ILE	10
1	A	136	ILE	9
1	A	97	LEU	9
1	A	19	LEU	8
1	A	34	LEU	8
1	A	137	LEU	7
1	A	40	LYS	7

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Mol	Chain	Res	Type	Models (Total)
1	A	86	PHE	7
1	A	72	THR	7
1	A	106	LYS	6
1	A	13	SER	6
1	A	17	LYS	6
1	A	102	PHE	6
1	A	35	ASP	6
1	A	75	LEU	6
1	A	85	LYS	6
1	A	89	LYS	5
1	A	53	LYS	5
1	A	132	ARG	5
1	A	11	TRP	5
1	A	104	LYS	5
1	A	74	LEU	5
1	A	50	GLU	5
1	A	33	LYS	5
1	A	135	LYS	5
1	A	128	VAL	4
1	A	108	TYR	4
1	A	6	LEU	4
1	A	107	ASP	4
1	A	59	LYS	4
1	A	57	VAL	4
1	A	115	ASN	4
1	A	7	GLU	4
1	A	82	GLN	4
1	A	139	LYS	4
1	A	79	LYS	4
1	A	42	ASP	4
1	A	129	CYS	3
1	A	63	ASP	3
1	A	101	GLU	3
1	A	26	LEU	3
1	A	41	VAL	3
1	A	93	PHE	3
1	A	9	ILE	3
1	A	69	LYS	3
1	A	130	GLN	3
1	A	24	LEU	3
1	A	98	TRP	3
1	A	84	ILE	3

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Mol	Chain	Res	Type	Models (Total)
1	A	68	LYS	3
1	A	27	TYR	2
1	A	65	CYS	2
1	A	91	GLN	2
1	A	36	ILE	2
1	A	5	VAL	2
1	A	77	CYS	2
1	A	92	GLU	2
1	A	100	LEU	2
1	A	81	ASP	2
1	A	14	SER	2
1	A	38	CYS	2
1	A	32	ASP	2
1	A	109	TYR	2
1	A	15	ASN	2
1	A	45	THR	2
1	A	60	ASP	1
1	A	138	MET	1
1	A	114	SER	1
1	A	96	ASN	1
1	A	56	MET	1
1	A	103	GLN	1
1	A	67	ILE	1
1	A	140	VAL	1
1	A	88	ILE	1
1	A	16	SER	1
1	A	123	ASN	1
1	A	76	ASN	1
1	A	142	GLN	1
1	A	3	SER	1
1	A	29	GLN	1
1	A	83	ASP	1
1	A	54	VAL	1
1	A	111	ILE	1
1	A	61	GLN	1
1	A	105	ASN	1
1	A	43	SER	1
1	A	112	SER	1

### 6.3.3 RNA ⓘ

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

The completeness of assignment taking into account all chemical shift lists is 75% for the well-defined parts and 76% for the entire structure.

### 7.1 Chemical shift list 1

File name: BMRB entry 7220

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	1515
Number of shifts mapped to atoms	1439
Number of unparsed shifts	0
Number of shifts with mapping errors	76
Number of shifts with mapping warnings	0
Number of shift outliers (ShiftChecker)	1

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

- Residue not found in structure. All 76 occurrences are reported below.

Chain	Res	Type	Atom	Shift Data		
				Value	Uncertainty	Ambiguity
A	150	THR	CB	70.29	0.1	1
A	151	ARG	CB	31.65	0.1	1
A	143	ASP	C	173.05	0.1	1
A	151	ARG	N	127.8	0.1	1
A	147	ALA	CA	53.26	0.1	1
A	147	ALA	HB1	1.41	0.05	1
A	151	ARG	HB3	1.85	0.05	2
A	146	SER	CA	59.11	0.1	1
A	144	ALA	CA	53.53	0.1	1
A	150	THR	N	116.49	0.1	1
A	148	GLY	HA3	3.97	0.05	2
A	143	ASP	HB3	2.65	0.05	2
A	143	ASP	CB	43.2	0.1	1
A	148	GLY	N	108.0	0.1	1

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Chain	Res	Type	Atom	Shift Data		
				Value	Uncertainty	Ambiguity
A	146	SER	CB	64.12	0.1	1
A	144	ALA	CB	18.75	0.1	1
A	144	ALA	HB3	0.95	0.05	1
A	146	SER	HB2	3.92	0.05	2
A	143	ASP	HA	4.51	0.05	1
A	150	THR	CA	62.42	0.1	1
A	146	SER	H	8.02	0.05	1
A	149	SER	HB2	3.88	0.05	2
A	150	THR	C	170.93	0.1	1
A	147	ALA	N	125.85	0.1	1
A	143	ASP	CA	53.84	0.1	1
A	148	GLY	C	171.53	0.1	1
A	145	SER	HA	4.3	0.05	1
A	149	SER	HA	4.52	0.05	1
A	146	SER	C	171.82	0.1	1
A	148	GLY	H	8.3	0.05	1
A	145	SER	HB3	3.87	0.05	2
A	145	SER	HB2	3.82	0.05	2
A	145	SER	N	114.28	0.1	1
A	147	ALA	HB2	1.41	0.05	1
A	149	SER	CB	64.4	0.1	1
A	150	THR	H	8.24	0.05	1
A	148	GLY	HA2	3.65	0.05	2
A	151	ARG	CG	30.0	0.1	1
A	151	ARG	H	7.62	0.05	1
A	149	SER	H	8.16	0.05	1
A	143	ASP	HB2	2.52	0.05	2
A	150	THR	HA	4.33	0.05	1
A	146	SER	N	117.48	0.1	1
A	144	ALA	N	126.94	0.1	1
A	145	SER	C	172.35	0.1	1
A	151	ARG	C	178.17	0.1	1
A	149	SER	N	115.64	0.1	1
A	147	ALA	HA	4.31	0.05	1
A	146	SER	HB3	3.86	0.05	2
A	144	ALA	HA	3.41	0.05	1
A	151	ARG	HA	4.2	0.05	1
A	151	ARG	HG2	1.6	0.05	2
A	149	SER	CA	58.8	0.1	1
A	145	SER	H	8.3	0.05	1
A	147	ALA	H	8.1	0.05	1

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Chain	Res	Type	Atom	Shift Data		
				Value	Uncertainty	Ambiguity
A	151	ARG	CD	44.69	0.1	1
A	145	SER	CB	63.94	0.1	1
A	148	GLY	CA	45.79	0.1	1
A	147	ALA	CB	19.4	0.1	1
A	151	ARG	HD2	3.18	0.05	2
A	144	ALA	HB1	0.95	0.05	1
A	143	ASP	H	8.51	0.05	1
A	147	ALA	C	175.52	0.1	1
A	150	THR	HG23	1.19	0.05	1
A	144	ALA	C	175.7	0.1	1
A	151	ARG	CA	58.04	0.1	1
A	150	THR	HG21	1.19	0.05	1
A	150	THR	HG22	1.19	0.05	1
A	145	SER	CA	59.84	0.1	1
A	144	ALA	H	8.27	0.05	1
A	151	ARG	HB2	1.705	0.05	2
A	149	SER	C	172.05	0.1	1
A	143	ASP	N	119.57	0.1	1
A	144	ALA	HB2	0.95	0.05	1
A	146	SER	HA	4.41	0.05	1
A	147	ALA	HB3	1.41	0.05	1

### 7.1.2 Chemical shift referencing [i](#)

The following table shows the suggested chemical shift referencing corrections.

Nucleus	# values	Correction $\pm$ precision, ppm	Suggested action
$^{13}\text{C}_\alpha$	146	$-0.41 \pm 0.23$	None needed ( $< 0.5$ ppm)
$^{13}\text{C}_\beta$	134	$-0.56 \pm 0.18$	Should be applied
$^{13}\text{C}'$	140	$3.10 \pm 0.16$	Should be applied
$^{15}\text{N}$	140	$-0.48 \pm 0.51$	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 75%, i.e. 1237 atoms were assigned a chemical shift out of a possible 1642. 9 out of 17 assigned methyl groups (LEU and VAL) were assigned stereospecifically.

	Total	$^1\text{H}$	$^{13}\text{C}$	$^{15}\text{N}$
Backbone	604/636 (95%)	241/253 (95%)	244/260 (94%)	119/123 (97%)

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	<b>Total</b>	<b><sup>1</sup>H</b>	<b><sup>13</sup>C</b>	<b><sup>15</sup>N</b>
Sidechain	633/881 (72%)	381/520 (73%)	239/323 (74%)	13/38 (34%)
Aromatic	0/125 (0%)	0/65 (0%)	0/58 (0%)	0/2 (0%)
Overall	1237/1642 (75%)	622/838 (74%)	483/641 (75%)	132/163 (81%)

The following table shows the completeness of the chemical shift assignments for the full structure. The overall completeness is 76%, i.e. 1358 atoms were assigned a chemical shift out of a possible 1777. 11 out of 20 assigned methyl groups (LEU and VAL) were assigned stereospecifically.

	<b>Total</b>	<b><sup>1</sup>H</b>	<b><sup>13</sup>C</b>	<b><sup>15</sup>N</b>
Backbone	664/696 (95%)	265/277 (96%)	268/284 (94%)	131/135 (97%)
Sidechain	694/948 (73%)	419/559 (75%)	261/349 (75%)	14/40 (35%)
Aromatic	0/133 (0%)	0/69 (0%)	0/62 (0%)	0/2 (0%)
Overall	1358/1777 (76%)	684/905 (76%)	529/695 (76%)	145/177 (82%)

#### 7.1.4 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	134	MET	HG2	0.45	4.23 – 0.63	-5.5

#### 7.1.5 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:

