Full wwPDB NMR Structure Validation Report (i)

Jun 22, 2024 – 09:45 PM EDT

PDB ID	:	6BUT
BMRB ID	:	27095
Title	:	Solution structure of full-length apo mammalian calmodulin bound to the IQ motif of the human voltage-gated sodium channel NaV1.2
Authors	:	Mahling, R.; Kilpatrick, A.M.; Shea, M.A.
Deposited on	:	2017-12-11

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 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:

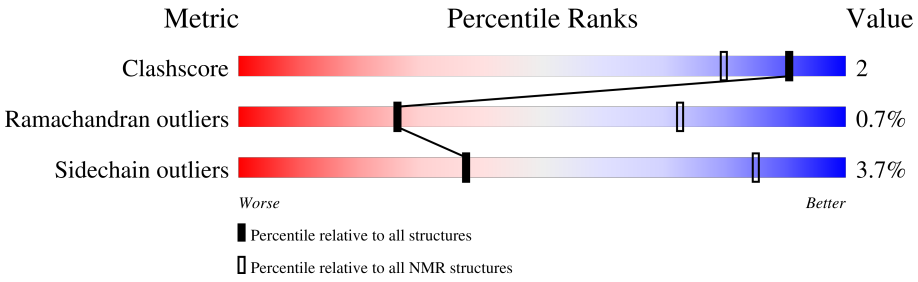
MolProbity	:	4.02b-467
Percentile statistics	:	20191225.v01 (using entries in the PDB archive December 25th 2019)
wwPDB-RCI	:	v_1n_11_5_13_A (Berjanski et al., 2005)
PANAV	:	Wang et al. (2010)
wwPDB-ShiftChecker	:	v1.2
Ideal geometry (proteins)	:	Engh & Huber (2001)
Ideal geometry (DNA, RNA)	:	Parkinson et al. (1996)
Validation Pipeline (wwPDB-VP)	:	2.37.1

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 89%.

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	148	74% 8% 18%
2	B	31	45% 10% 45%

2 Ensemble composition and analysis (i)

This entry contains 20 models. Model 18 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 average pairwise backbone rmsd*.

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:6-A:20, A:24-A:73 (65)	0.34	5
2	A:82-A:93, A:98-A:129, A:134-A:146, B:1904-B:1920 (74)	0.24	18

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 1 single-model cluster was found.

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

3 Entry composition (i)

There are 2 unique types of molecules in this entry. The entry contains 2813 atoms, of which 1388 are hydrogens and 0 are deuteriums.

- Molecule 1 is a protein called Calmodulin-1.

Mol	Chain	Residues	Atoms						Trace
			Total	C	H	N	O	S	
1	A	148	2262	714	1096	188	255	9	0

- Molecule 2 is a protein called Sodium channel protein type 2 subunit alpha.

Mol	Chain	Residues	Atoms					Trace
			Total	C	H	N	O	
2	B	31	551	164	292	52	43	0

There are 4 discrepancies between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	Comment	Reference
B	-4	GLY	-	expression tag	UNP Q99250
B	-3	PRO	-	expression tag	UNP Q99250
B	-2	GLY	-	expression tag	UNP Q99250
B	-1	SER	-	expression tag	UNP Q99250

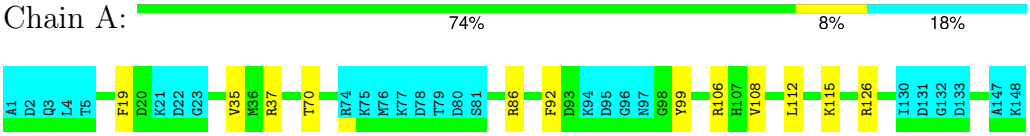
4 Residue-property plots (i)

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.

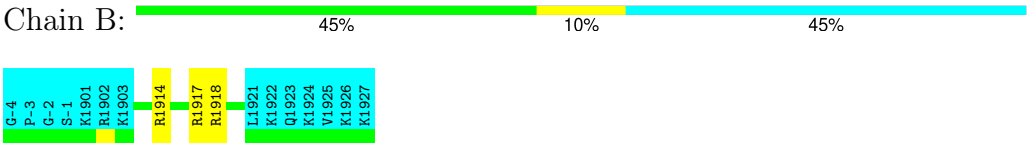
• Molecule 1: Calmodulin-1

Chain A:



• Molecule 2: Sodium channel protein type 2 subunit alpha

Chain B:



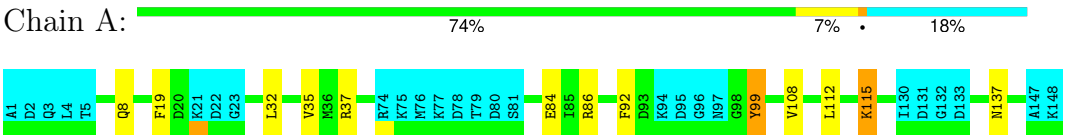
4.2 Scores per residue for each member of the ensemble

Colouring as in section 4.1 above.

4.2.1 Score per residue for model 1

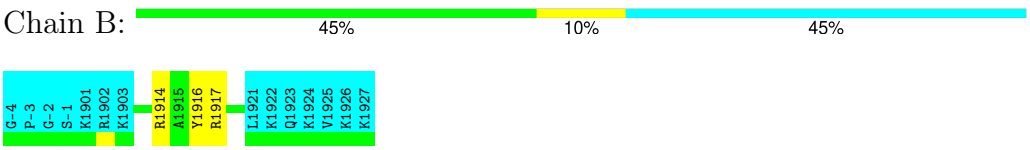
• Molecule 1: Calmodulin-1

Chain A:



• Molecule 2: Sodium channel protein type 2 subunit alpha

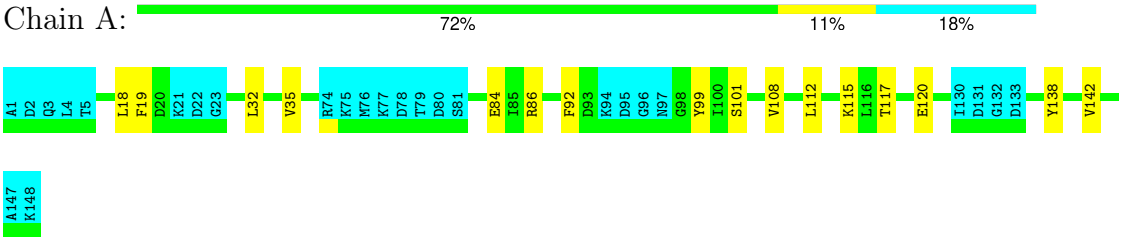
Chain B:



4.2.2 Score per residue for model 2

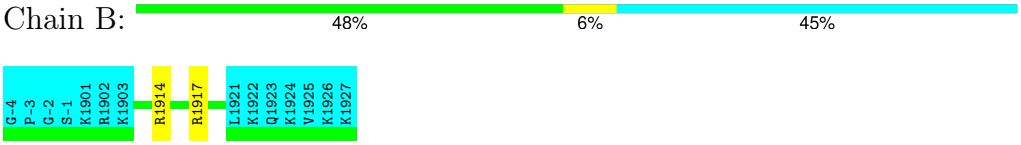
• Molecule 1: Calmodulin-1

Chain A:



• Molecule 2: Sodium channel protein type 2 subunit alpha

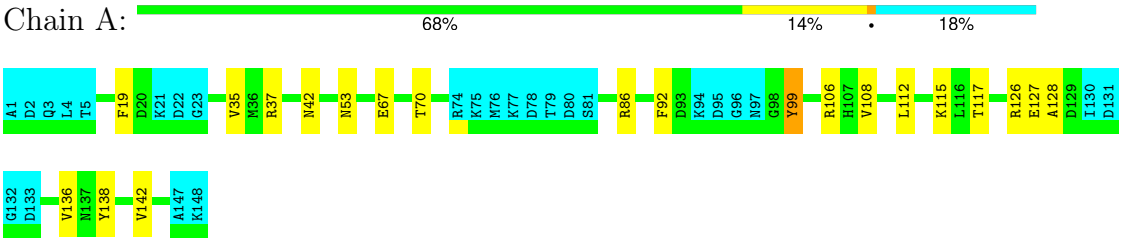
Chain B:



4.2.3 Score per residue for model 3

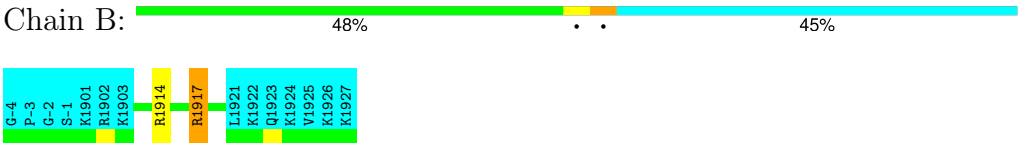
• Molecule 1: Calmodulin-1

Chain A:



• Molecule 2: Sodium channel protein type 2 subunit alpha

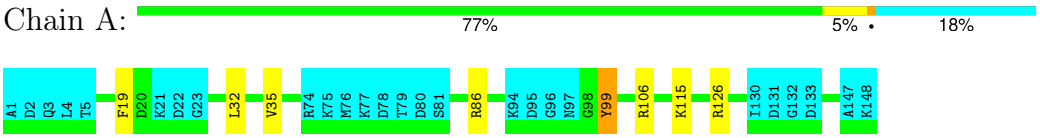
Chain B:



4.2.4 Score per residue for model 4

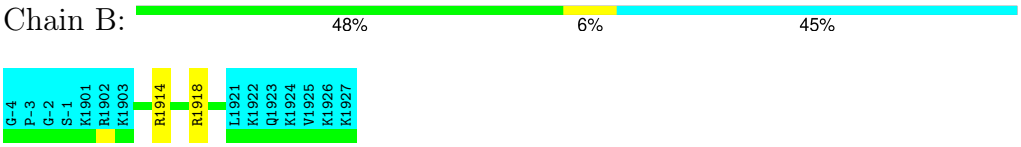
• Molecule 1: Calmodulin-1

Chain A:



• Molecule 2: Sodium channel protein type 2 subunit alpha

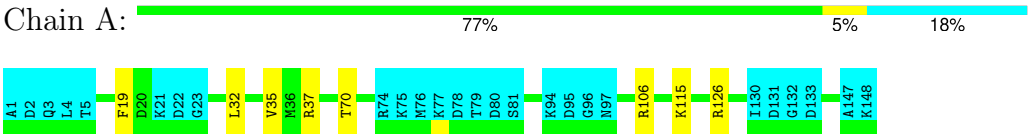
Chain B:



4.2.5 Score per residue for model 5

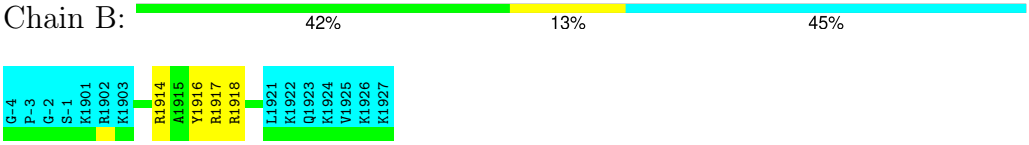
• Molecule 1: Calmodulin-1

Chain A:



• Molecule 2: Sodium channel protein type 2 subunit alpha

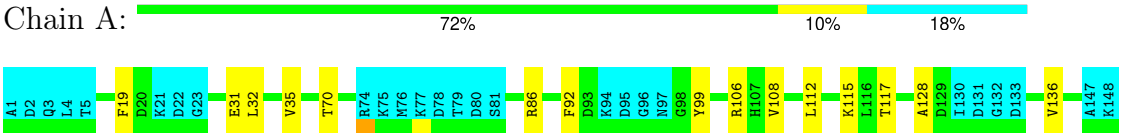
Chain B:



4.2.6 Score per residue for model 6

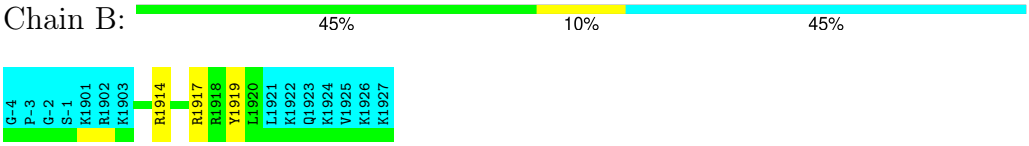
• Molecule 1: Calmodulin-1

Chain A:



• Molecule 2: Sodium channel protein type 2 subunit alpha

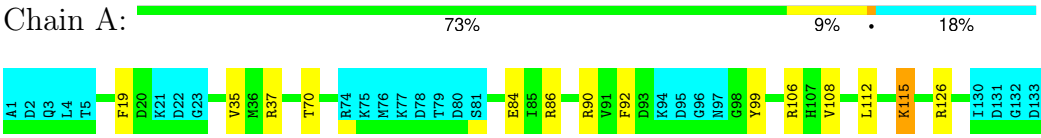
Chain B:



4.2.7 Score per residue for model 7

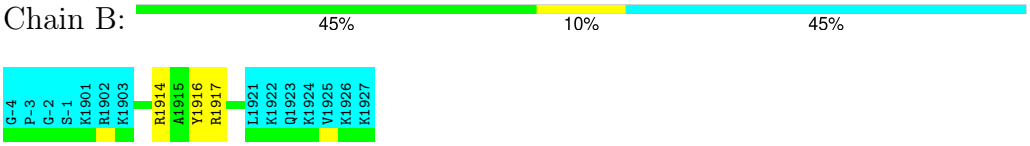
• Molecule 1: Calmodulin-1

Chain A:



• Molecule 2: Sodium channel protein type 2 subunit alpha

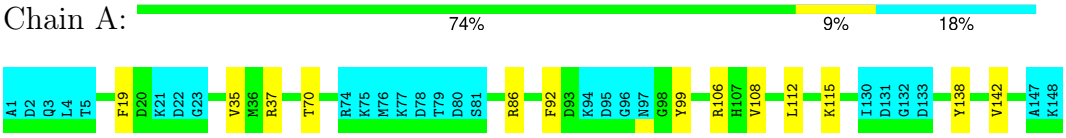
Chain B:



4.2.8 Score per residue for model 8

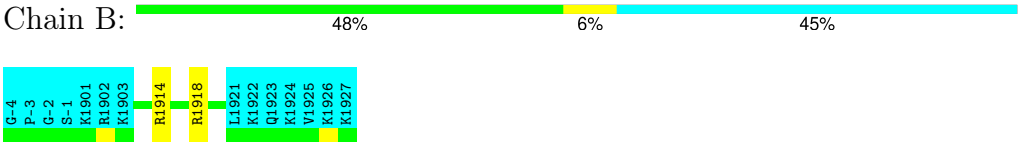
• Molecule 1: Calmodulin-1

Chain A:



• Molecule 2: Sodium channel protein type 2 subunit alpha

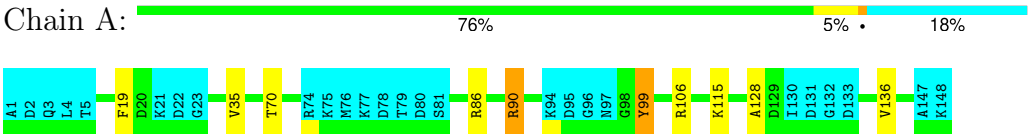
Chain B:



4.2.9 Score per residue for model 9

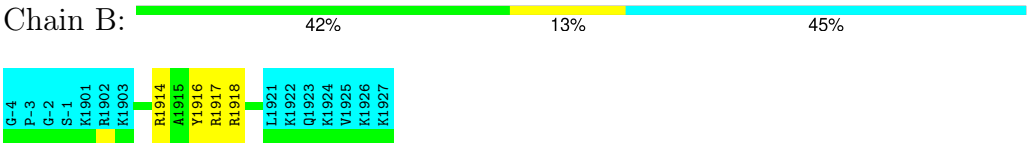
• Molecule 1: Calmodulin-1

Chain A:



• Molecule 2: Sodium channel protein type 2 subunit alpha

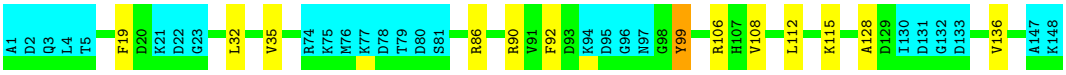
Chain B:



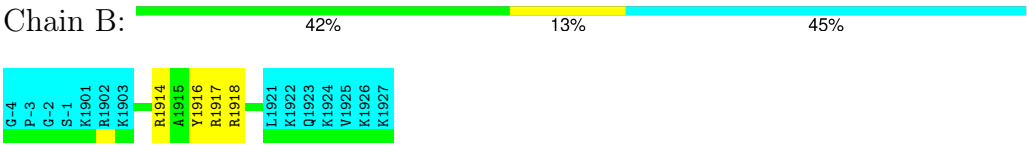
4.2.10 Score per residue for model 10

• Molecule 1: Calmodulin-1

Chain A:

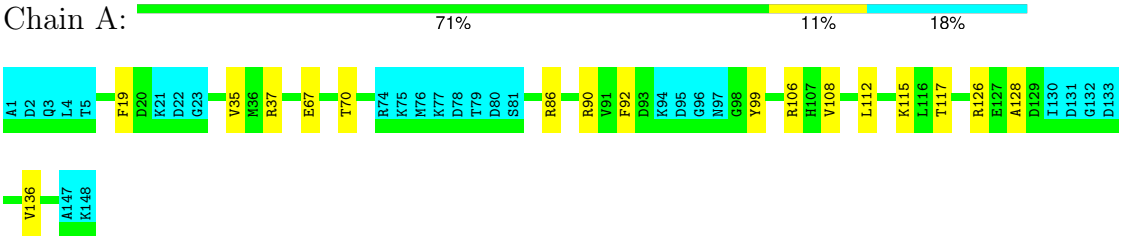


• Molecule 2: Sodium channel protein type 2 subunit alpha

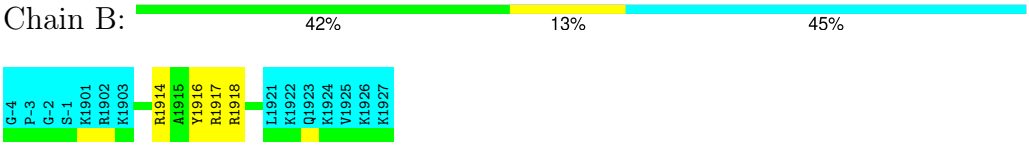


4.2.11 Score per residue for model 11

• Molecule 1: Calmodulin-1

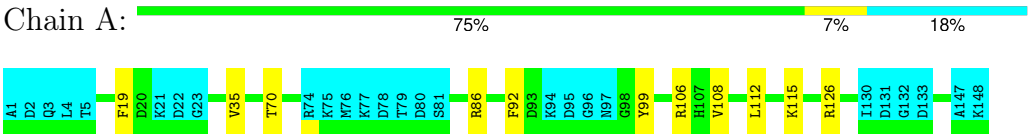


• Molecule 2: Sodium channel protein type 2 subunit alpha



4.2.12 Score per residue for model 12

• Molecule 1: Calmodulin-1

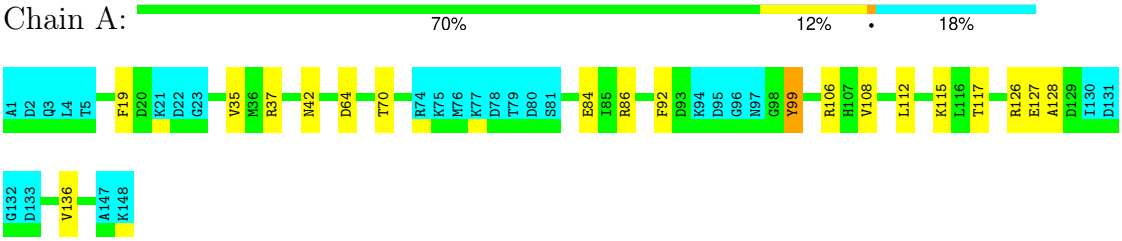


• Molecule 2: Sodium channel protein type 2 subunit alpha

4.2.13 Score per residue for model 13

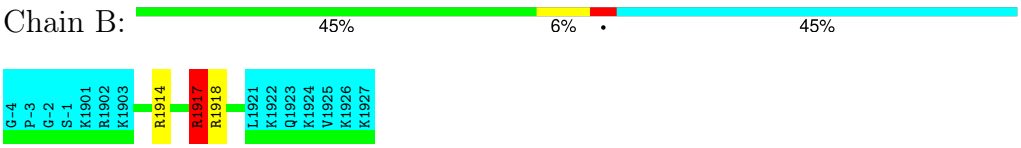
• Molecule 1: Calmodulin-1

Chain A:



• Molecule 2: Sodium channel protein type 2 subunit alpha

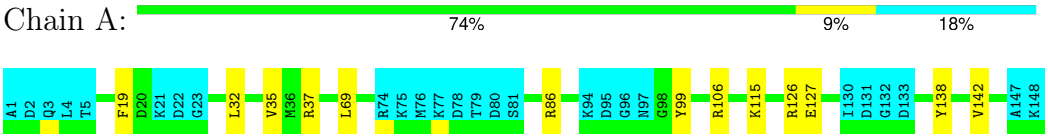
Chain B:



4.2.14 Score per residue for model 14

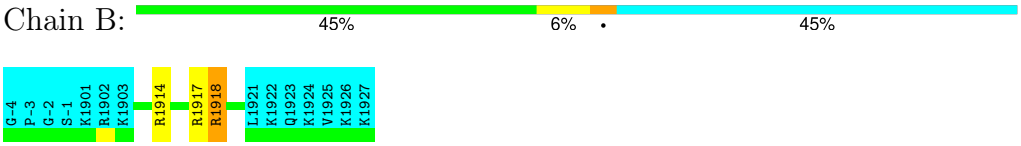
• Molecule 1: Calmodulin-1

Chain A:



• Molecule 2: Sodium channel protein type 2 subunit alpha

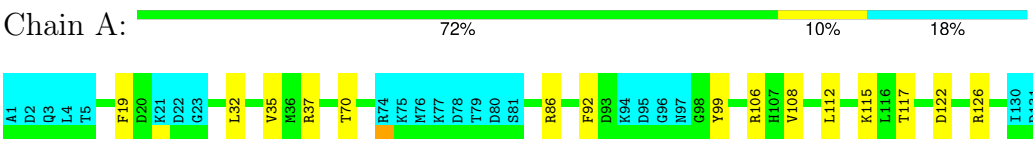
Chain B:



4.2.15 Score per residue for model 15

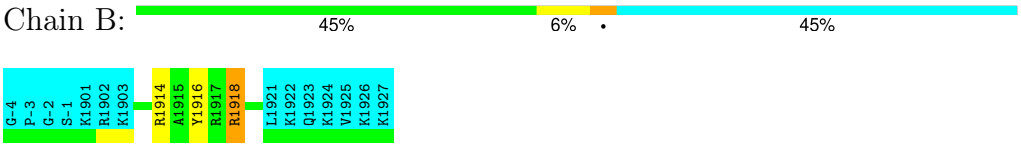
• Molecule 1: Calmodulin-1

Chain A:



• Molecule 2: Sodium channel protein type 2 subunit alpha

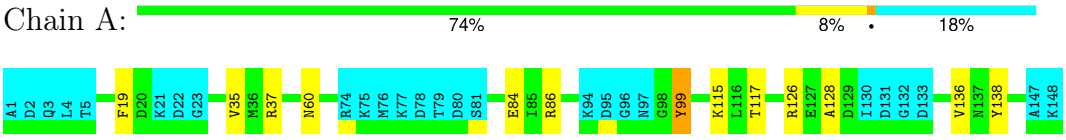
Chain B:



4.2.16 Score per residue for model 16

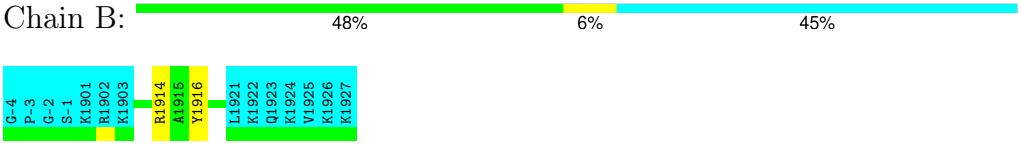
• Molecule 1: Calmodulin-1

Chain A:



• Molecule 2: Sodium channel protein type 2 subunit alpha

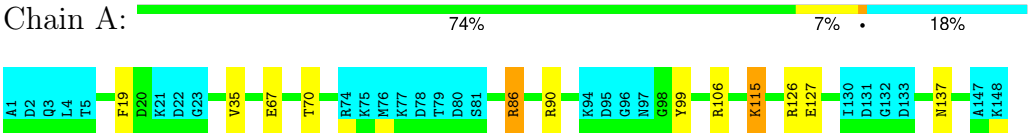
Chain B:



4.2.17 Score per residue for model 17

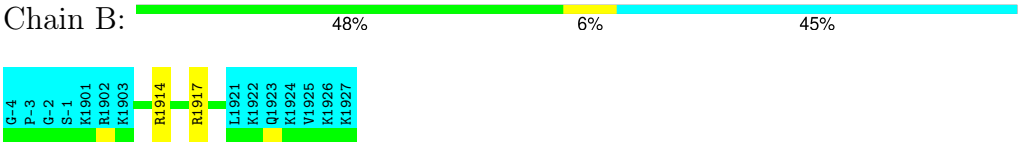
• Molecule 1: Calmodulin-1

Chain A:



• Molecule 2: Sodium channel protein type 2 subunit alpha

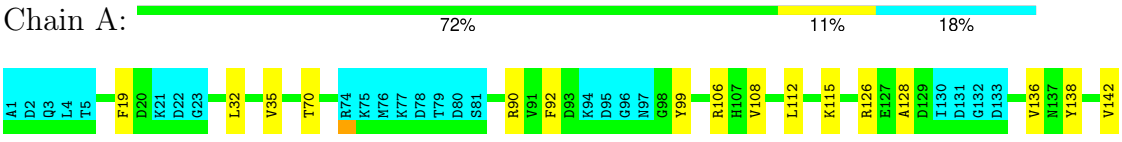
Chain B:



4.2.18 Score per residue for model 18 (medoid)

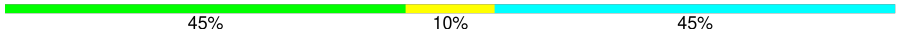
• Molecule 1: Calmodulin-1

Chain A:



• Molecule 2: Sodium channel protein type 2 subunit alpha

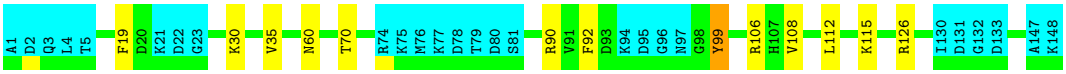
Chain B:



4.2.19 Score per residue for model 19

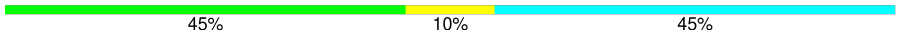
• Molecule 1: Calmodulin-1

Chain A:



• Molecule 2: Sodium channel protein type 2 subunit alpha

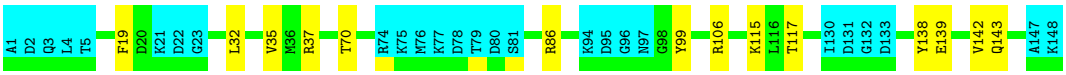
Chain B:



4.2.20 Score per residue for model 20

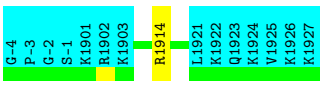
• Molecule 1: Calmodulin-1

Chain A:



• Molecule 2: Sodium channel protein type 2 subunit alpha

Chain B:



5 Refinement protocol and experimental data overview

The models were refined using the following method: *molecular dynamics*.

Of the 40 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
CYANA	structure calculation	2.1
Amber	refinement	AMBERTOOLS 17

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	4
Total number of shifts	2115
Number of shifts mapped to atoms	2115
Number of unparsed shifts	0
Number of shifts with mapping errors	0
Number of shifts with mapping warnings	0
Assignment completeness (well-defined parts)	89%

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	0.71±0.00	0±0/983 (0.0± 0.0%)	1.02±0.02	3±1/1325 (0.3± 0.1%)
2	B	0.81±0.02	0±0/150 (0.0± 0.0%)	1.30±0.07	3±1/202 (1.5± 0.5%)
All	All	0.72	0/22660 (0.0%)	1.06	126/30540 (0.4%)

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	0.6±0.6
2	B	0.0±0.0	0.1±0.2
All	All	0	13

There are no bond-length outliers.

All 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	106	ARG	NE-CZ-NH1	9.92	125.26	120.30	9	17
2	B	1917	ARG	NE-CZ-NH1	9.11	124.85	120.30	17	14
1	A	126	ARG	NE-CZ-NH1	9.06	124.83	120.30	11	13
1	A	86	ARG	NE-CZ-NH1	9.05	124.82	120.30	13	17
2	B	1914	ARG	NE-CZ-NH1	8.91	124.75	120.30	3	20
1	A	37	ARG	NE-CZ-NH1	8.42	124.51	120.30	20	11
2	B	1918	ARG	NE-CZ-NH1	7.84	124.22	120.30	5	9
1	A	90	ARG	NE-CZ-NH1	6.59	123.59	120.30	9	7
1	A	90	ARG	NE-CZ-NH2	-6.30	117.15	120.30	9	1
2	B	1916	TYR	CB-CG-CD1	-5.60	117.64	121.00	16	6
2	B	1916	TYR	CB-CG-CD2	-5.45	117.73	121.00	9	3
2	B	1914	ARG	NE-CZ-NH2	-5.36	117.62	120.30	19	5
2	B	1917	ARG	NE-CZ-NH2	-5.31	117.64	120.30	13	1

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Mol	Chain	Res	Type	Atoms	Z	Observed(^{o})	Ideal(^{o})	Models	
								Worst	Total
2	B	1919	TYR	CB-CG-CD1	-5.20	117.88	121.00	6	1
1	A	126	ARG	NE-CZ-NH2	-5.11	117.74	120.30	14	1

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	99	TYR	Sidechain	11
2	B	1917	ARG	Sidechain	1
1	A	138	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	970	905	905	4±2
All	All	22360	21160	21160	71

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 2.

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

Atom-1	Atom-2	Clash(Å)	Distance(Å)	Models	
				Worst	Total
1:A:108:VAL:HG23	1:A:112:LEU:HD12	0.72	1.62	13	12
1:A:92:PHE:CZ	1:A:108:VAL:HG21	0.60	2.32	15	12
1:A:128:ALA:HB1	1:A:136:VAL:HG13	0.57	1.77	16	8
1:A:19:PHE:CD1	1:A:35:VAL:HG12	0.54	2.37	12	5
1:A:19:PHE:CD2	1:A:35:VAL:HG12	0.53	2.39	7	5
1:A:138:TYR:O	1:A:142:VAL:HG23	0.53	2.04	8	6
1:A:92:PHE:CE2	1:A:108:VAL:HG21	0.51	2.41	19	8
1:A:92:PHE:CE1	1:A:108:VAL:HG21	0.49	2.43	15	3
1:A:19:PHE:CD2	1:A:35:VAL:HG22	0.47	2.43	15	7
1:A:19:PHE:CD1	1:A:35:VAL:HG22	0.45	2.46	10	3
1:A:92:PHE:CZ	1:A:108:VAL:HG11	0.42	2.50	1	1

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Atom-1	Atom-2	Clash(Å)	Distance(Å)	Models	
				Worst	Total
1:A:108:VAL:HG13	1:A:112:LEU:HD12	0.41	1.92	1	1

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	122/148 (82%)	118±2 (96±1%)	3±2 (3±1%)	1±0 (1±0%)	24	71
2	B	17/31 (55%)	17±0 (100±0%)	0±0 (0±0%)	0±0 (0±0%)	100	100
All	All	2780/3580 (78%)	2694 (97%)	66 (2%)	20 (1%)	26	73

All 1 unique Ramachandran outliers are listed below.

Mol	Chain	Res	Type	Models (Total)
1	A	115	LYS	20

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	105/126 (83%)	101±2 (96±2%)	4±2 (4±2%)	35	83
2	B	15/27 (56%)	15±0 (98±3%)	0±0 (2±3%)	62	94
All	All	2400/3060 (78%)	2312 (96%)	88 (4%)	37	85

All 27 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	99	TYR	16

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Mol	Chain	Res	Type	Models (Total)
1	A	70	THR	14
1	A	32	LEU	10
1	A	117	THR	8
1	A	84	GLU	5
1	A	127	GLU	4
1	A	115	LYS	3
1	A	67	GLU	3
2	B	1918	ARG	3
1	A	137	ASN	2
1	A	42	ASN	2
2	B	1917	ARG	2
1	A	60	ASN	2
1	A	8	GLN	1
1	A	18	LEU	1
1	A	101	SER	1
1	A	120	GLU	1
1	A	53	ASN	1
1	A	31	GLU	1
1	A	90	ARG	1
1	A	64	ASP	1
1	A	69	LEU	1
1	A	122	ASP	1
1	A	86	ARG	1
1	A	30	LYS	1
1	A	139	GLU	1
1	A	143	GLN	1

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 89% for the well-defined parts and 86% 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	1705
Number of shifts mapped to atoms	1705
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

The following table shows the suggested chemical shift referencing corrections.

Nucleus	# values	Correction ± precision, *ppm*	Suggested action
$^{13}C_{\alpha}$	148	-0.28 ± 0.05	None needed (< 0.5 ppm)
$^{13}C_{\beta}$	137	0.06 ± 0.09	None needed (< 0.5 ppm)
$^{13}C'$	144	-0.35 ± 0.07	None needed (< 0.5 ppm)
^{15}N	141	0.56 ± 0.31	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 77%, i.e. 1452 atoms were assigned a chemical shift out of a possible 1891. 0 out of 17 assigned methyl groups (LEU and VAL) were assigned stereospecifically.

	Total	^{1}H	^{13}C	^{15}N
Backbone	609/699 (87%)	248/284 (87%)	243/278 (87%)	118/137 (86%)
Sidechain	748/1069 (70%)	493/688 (72%)	248/342 (73%)	7/39 (18%)

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	Total	^{1}H	^{13}C	^{15}N
Aromatic	95/123 (77%)	49/60 (82%)	46/62 (74%)	0/1 (0%)
Overall	1452/1891 (77%)	790/1032 (77%)	537/682 (79%)	125/177 (71%)

The following table shows the completeness of the chemical shift assignments for the full structure. The overall completeness is 70%, i.e. 1705 atoms were assigned a chemical shift out of a possible 2422. 0 out of 20 assigned methyl groups (LEU and VAL) were assigned stereospecifically.

	Total	^{1}H	^{13}C	^{15}N
Backbone	729/902 (81%)	296/368 (80%)	292/358 (82%)	141/176 (80%)
Sidechain	881/1397 (63%)	576/893 (65%)	296/445 (67%)	9/59 (15%)
Aromatic	95/123 (77%)	49/60 (82%)	46/62 (74%)	0/1 (0%)
Overall	1705/2422 (70%)	921/1321 (70%)	634/865 (73%)	150/236 (64%)

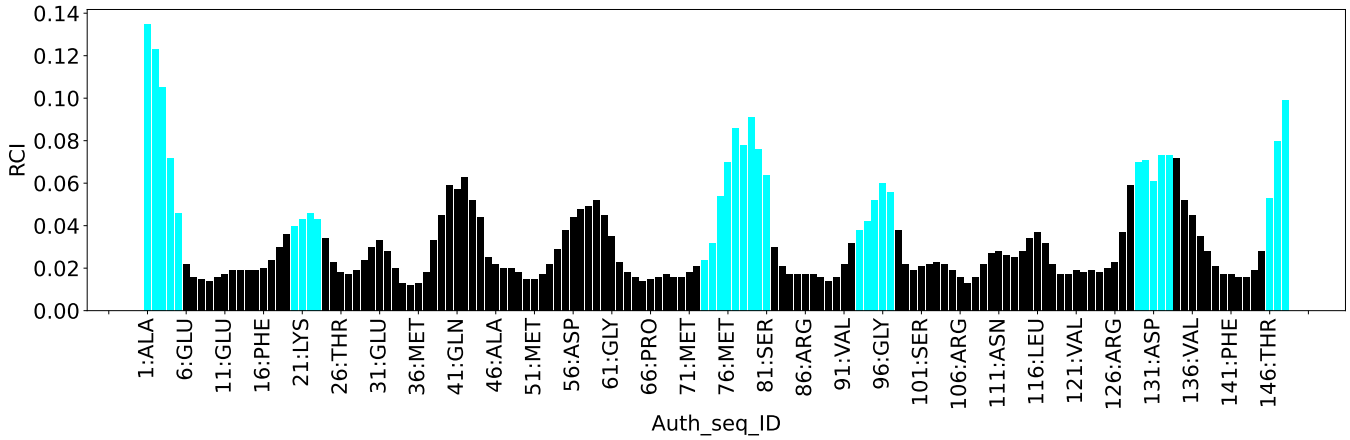
7.1.4 Statistically unusual chemical shifts (i)

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:



7.2 Chemical shift list 2

File name: working_cs.cif

Chemical shift list name: *assigned_chem_shift_list_2*

7.2.1 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	368
Number of shifts mapped to atoms	368
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

7.2.2 Chemical shift referencing

The following table shows the suggested chemical shift referencing corrections.

Nucleus	# values	Correction ± precision, *ppm*	Suggested action
$^{13}C_{\alpha}$	30	-0.80 ± 0.19	Should be checked
$^{13}C_{\beta}$	28	0.13 ± 0.08	None needed (< 0.5 ppm)
$^{13}C'$	29	-0.47 ± 0.18	None needed (< 0.5 ppm)
^{15}N	28	-0.05 ± 0.31	None needed (< 0.5 ppm)

7.2.3 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 12%, i.e. 223 atoms were assigned a chemical shift out of a possible 1891. 0 out of 17 assigned methyl groups (LEU and VAL) were assigned stereospecifically.

	Total	^{1}H	^{13}C	^{15}N
Backbone	85/699 (12%)	34/284 (12%)	34/278 (12%)	17/137 (12%)
Sidechain	122/1069 (11%)	82/688 (12%)	40/342 (12%)	0/39 (0%)
Aromatic	16/123 (13%)	8/60 (13%)	8/62 (13%)	0/1 (0%)
Overall	223/1891 (12%)	124/1032 (12%)	82/682 (12%)	17/177 (10%)

The following table shows the completeness of the chemical shift assignments for the full structure. The overall completeness is 15%, i.e. 368 atoms were assigned a chemical shift out of a possible 2422. 0 out of 20 assigned methyl groups (LEU and VAL) were assigned stereospecifically.

	Total	^{1}H	^{13}C	^{15}N
Backbone	144/902 (16%)	57/368 (15%)	59/358 (16%)	28/176 (16%)
Sidechain	208/1397 (15%)	133/893 (15%)	75/445 (17%)	0/59 (0%)
Aromatic	16/123 (13%)	8/60 (13%)	8/62 (13%)	0/1 (0%)
Overall	368/2422 (15%)	198/1321 (15%)	142/865 (16%)	28/236 (12%)

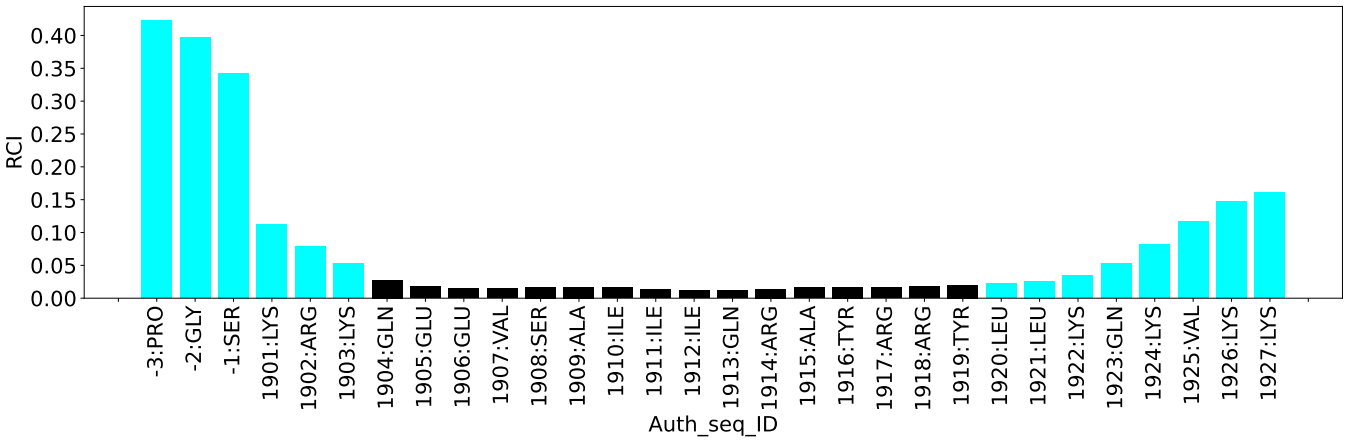
7.2.4 Statistically unusual chemical shifts (i)

There are no statistically unusual chemical shifts.

7.2.5 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 B:



7.3 Chemical shift list 3

File name: working_cs.cif

Chemical shift list name: *assigned_chem_shift_list_3*

7.3.1 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	19
Number of shifts mapped to atoms	19
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

7.3.2 Chemical shift referencing (i)

No chemical shift referencing corrections were calculated (not enough data).

7.3.3 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 0%, i.e. 0 atoms were assigned a chemical shift out of a possible 1891. 0 out of 17 assigned methyl groups (LEU and VAL) were assigned stereospecifically.

	Total	^{1}H	^{13}C	^{15}N
Backbone	0/699 (0%)	0/284 (0%)	0/278 (0%)	0/137 (0%)
Sidechain	0/1069 (0%)	0/688 (0%)	0/342 (0%)	0/39 (0%)
Aromatic	0/123 (0%)	0/60 (0%)	0/62 (0%)	0/1 (0%)
Overall	0/1891 (0%)	0/1032 (0%)	0/682 (0%)	0/177 (0%)

The following table shows the completeness of the chemical shift assignments for the full structure. The overall completeness is 1%, i.e. 19 atoms were assigned a chemical shift out of a possible 2422. 0 out of 20 assigned methyl groups (LEU and VAL) were assigned stereospecifically.

	Total	^{1}H	^{13}C	^{15}N
Backbone	15/902 (2%)	4/368 (1%)	7/358 (2%)	4/176 (2%)
Sidechain	4/1397 (0%)	0/893 (0%)	4/445 (1%)	0/59 (0%)
Aromatic	0/123 (0%)	0/60 (0%)	0/62 (0%)	0/1 (0%)
Overall	19/2422 (1%)	4/1321 (0%)	11/865 (1%)	4/236 (2%)

7.3.4 Statistically unusual chemical shifts (i)

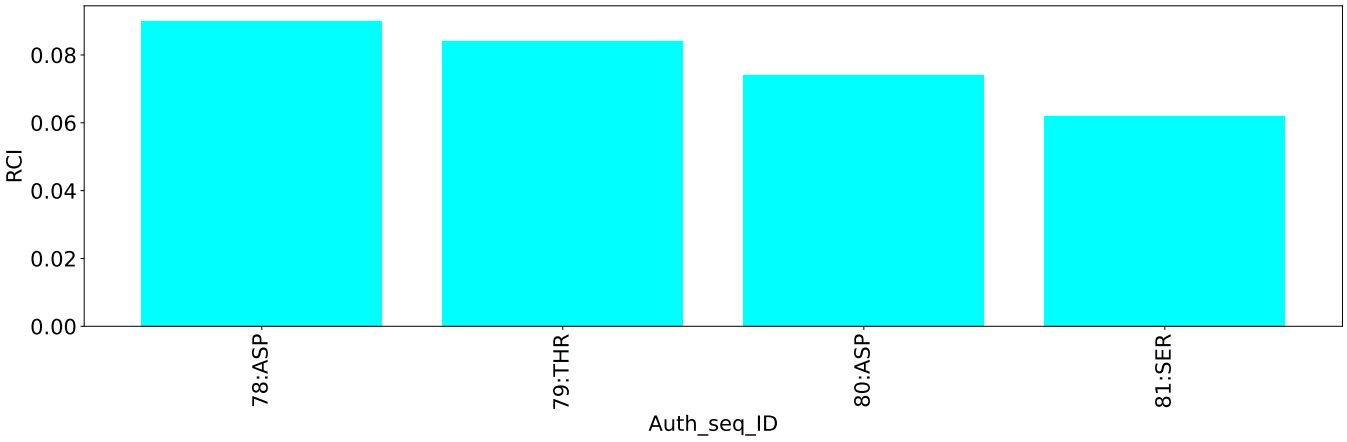
There are no statistically unusual chemical shifts.

7.3.5 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:



7.4 Chemical shift list 4

File name: working_cs.cif

Chemical shift list name: *assigned_chem_shift_list_4*

7.4.1 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	23
Number of shifts mapped to atoms	23
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

7.4.2 Chemical shift referencing

No chemical shift referencing corrections were calculated (not enough data).

7.4.3 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 1%, i.e. 18 atoms were assigned a chemical shift out of a possible 1891. 0 out of 17 assigned methyl groups (LEU and VAL) were assigned stereospecifically.

	Total	^{1}H	^{13}C	^{15}N
Backbone	14/699 (2%)	4/284 (1%)	6/278 (2%)	4/137 (3%)
Sidechain	4/1069 (0%)	0/688 (0%)	4/342 (1%)	0/39 (0%)
Aromatic	0/123 (0%)	0/60 (0%)	0/62 (0%)	0/1 (0%)
Overall	18/1891 (1%)	4/1032 (0%)	10/682 (1%)	4/177 (2%)

The following table shows the completeness of the chemical shift assignments for the full structure. The overall completeness is 1%, i.e. 23 atoms were assigned a chemical shift out of a possible 2422. 0 out of 20 assigned methyl groups (LEU and VAL) were assigned stereospecifically.

	Total	^{1}H	^{13}C	^{15}N
Backbone	18/902 (2%)	5/368 (1%)	8/358 (2%)	5/176 (3%)
Sidechain	5/1397 (0%)	0/893 (0%)	5/445 (1%)	0/59 (0%)
Aromatic	0/123 (0%)	0/60 (0%)	0/62 (0%)	0/1 (0%)
Overall	23/2422 (1%)	5/1321 (0%)	13/865 (2%)	5/236 (2%)

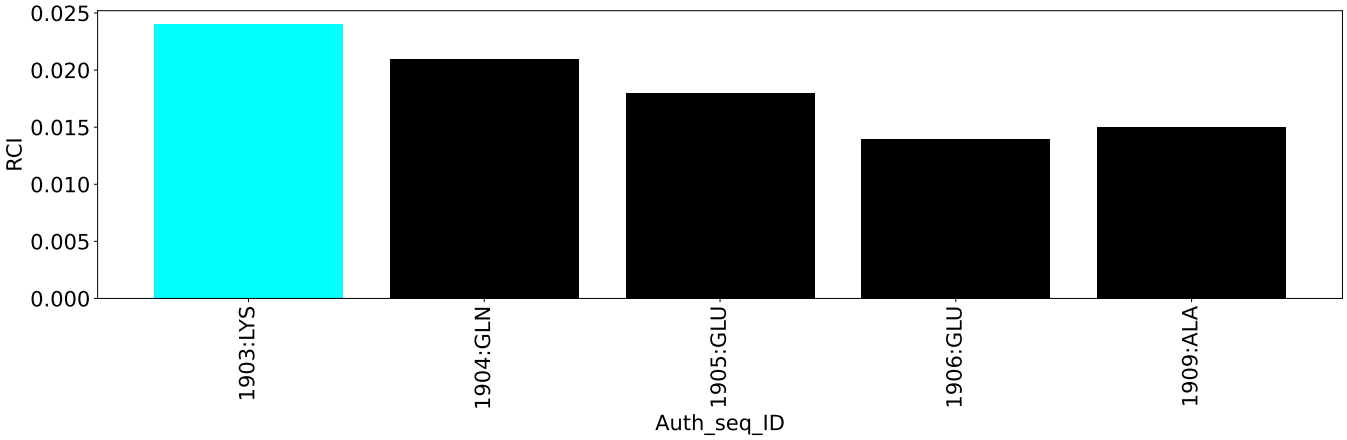
7.4.4 Statistically unusual chemical shifts

There are no statistically unusual chemical shifts.

7.4.5 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.If well-defined core and ill-defined regions are not identified then it is shown as gray bars.

Random coil index (RCI) for chain B:


0.025
0.020
0.015
0.010
0.005
0.000
RCI
1903:LYS
1904:GLN
1905:GLU
1906:GLU
1909:ALA
Auth_seq_ID