



wwPDB NMR Structure Validation Summary Report ⓘ

May 28, 2020 – 11:28 pm BST

PDB ID : 2LSP
Title : solution structures of BRD4 second bromodomain with NF-kB-K310ac peptide
Authors : Zhang, G.; Liu, R.; Zhong, Y.; Plotnikov, A.N.; Zhang, W.; Rusinova, E.;
Gerona-Nevarro, G.; Moshkina, N.; Joshua, J.; Chuang, P.Y.; Ohlmeyer, M.;
He, J.; Zhou, M.-M.
Deposited on : 2012-05-03

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 ⓘ symbol.

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 : 1.8.5 (274361), CSD as541be (2020)
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

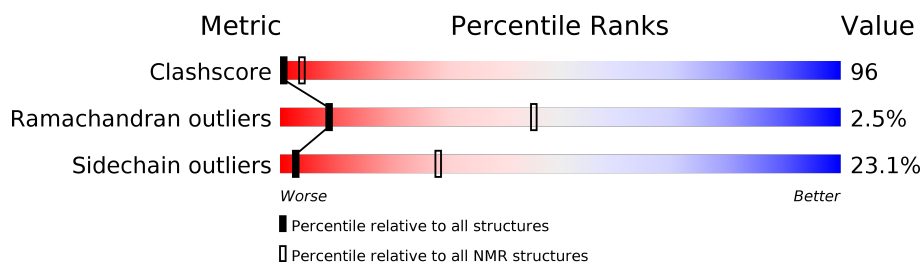
1 Overall quality at a glance

The following experimental techniques were used to determine the structure:

SOLUTION NMR

The overall completeness of chemical shifts assignment is 82%.

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 $\leq 5\%$

Mol	Chain	Length	Quality of chain
1	A	13	100%
2	B	128	16% 45% 18% 20%

2 Ensemble composition and analysis ⓘ

This entry contains 20 models. Model 17 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	B:353-B:454 (102)	0.10	17

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 4 single-model clusters were found.

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

3 Entry composition [i](#)

There are 2 unique types of molecules in this entry. The entry contains 2294 atoms, of which 1141 are hydrogens and 0 are deuteriums.

- Molecule 1 is a protein called NF-kB-K310ac peptide.

Mol	Chain	Residues	Atoms						Trace
1	A	13	Total	C	H	N	O	S	0
			239	74	123	19	22	1	

- Molecule 2 is a protein called Bromodomain-containing protein 4.

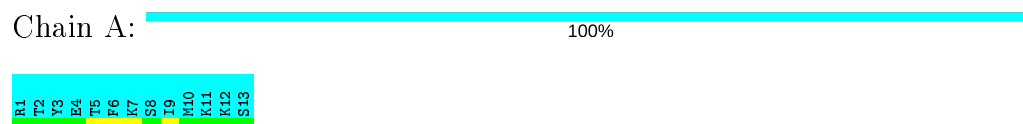
Mol	Chain	Residues	Atoms						Trace
2	B	128	Total	C	H	N	O	S	0
			2055	658	1018	175	193	11	

4 Residue-property plots

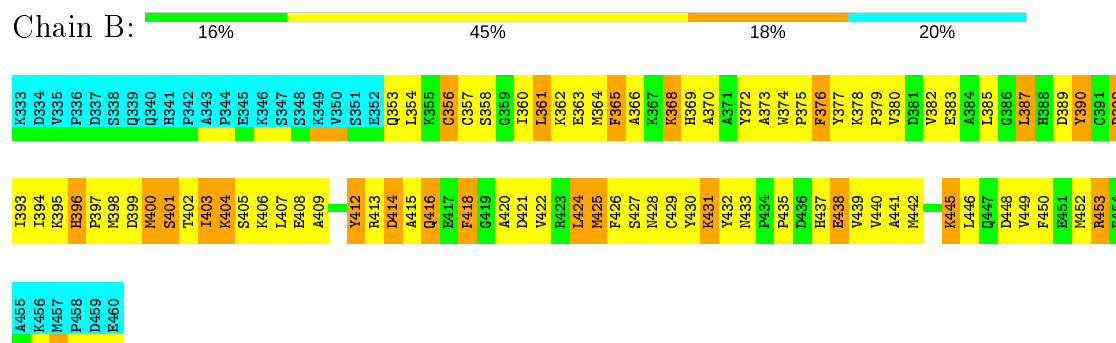
4.1 Average score per residue in the NMR ensemble

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: NF-kB-K310ac peptide



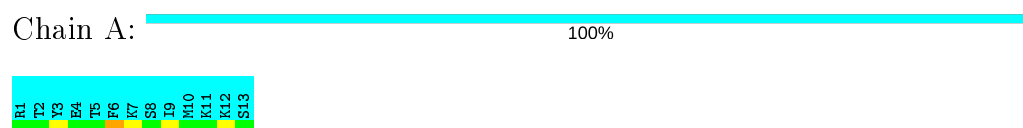
- Molecule 2: Bromodomain-containing protein 4



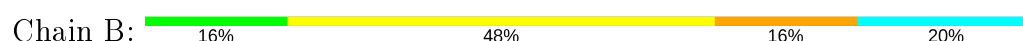
4.2 Residue scores for the representative (medoid) model from the NMR ensemble

The representative model is number 17. Colouring as in section 4.1 above.

- Molecule 1: NF-kB-K310ac peptide



- Molecule 2: Bromodomain-containing protein 4



I393	I394	I395	I396	I397	I398	I399	I400	I401	I402	I403	I404	I405	I406	I407	I408	I409	I410	I411	I412	I413	I414	I415	I416	I417	I418	I419	I420	I421	I422	I423	I424	I425	I426	I427	I428	I429	I430	I431	I432	I433	I434	I435	I436	I437	I438	I439	I440	I441	I442	I443	I444	I445	I446	I447	I448	I449	I450	I451	I452	I453	I454
K393	K394	K395	K396	K397	K398	K399	K400	K401	K402	K403	K404	K405	K406	K407	K408	K409	K410	K411	K412	K413	K414	K415	K416	K417	K418	K419	K420	K421	K422	K423	K424	K425	K426	K427	K428	K429	K430	K431	K432	K433	K434	K435	K436	K437	K438	K439	K440	K441	K442	K443	K444	K445	K446	K447	K448	K449	K450	K451	K452	K453	K454
L393	L394	L395	L396	L397	L398	L399	L400	L401	L402	L403	L404	L405	L406	L407	L408	L409	L410	L411	L412	L413	L414	L415	L416	L417	L418	L419	L420	L421	L422	L423	L424	L425	L426	L427	L428	L429	L430	L431	L432	L433	L434	L435	L436	L437	L438	L439	L440	L441	L442	L443	L444	L445	L446	L447	L448	L449	L450	L451	L452	L453	L454
M393	M394	M395	M396	M397	M398	M399	M400	M401	M402	M403	M404	M405	M406	M407	M408	M409	M410	M411	M412	M413	M414	M415	M416	M417	M418	M419	M420	M421	M422	M423	M424	M425	M426	M427	M428	M429	M430	M431	M432	M433	M434	M435	M436	M437	M438	M439	M440	M441	M442	M443	M444	M445	M446	M447	M448	M449	M450	M451	M452	M453	M454
N393	N394	N395	N396	N397	N398	N399	N400	N401	N402	N403	N404	N405	N406	N407	N408	N409	N410	N411	N412	N413	N414	N415	N416	N417	N418	N419	N420	N421	N422	N423	N424	N425	N426	N427	N428	N429	N430	N431	N432	N433	N434	N435	N436	N437	N438	N439	N440	N441	N442	N443	N444	N445	N446	N447	N448	N449	N450	N451	N452	N453	N454
O393	O394	O395	O396	O397	O398	O399	O400	O401	O402	O403	O404	O405	O406	O407	O408	O409	O410	O411	O412	O413	O414	O415	O416	O417	O418	O419	O420	O421	O422	O423	O424	O425	O426	O427	O428	O429	O430	O431	O432	O433	O434	O435	O436	O437	O438	O439	O440	O441	O442	O443	O444	O445	O446	O447	O448	O449	O450	O451	O452	O453	O454
P393	P394	P395	P396	P397	P398	P399	P400	P401	P402	P403	P404	P405	P406	P407	P408	P409	P410	P411	P412	P413	P414	P415	P416	P417	P418	P419	P420	P421	P422	P423	P424	P425	P426	P427	P428	P429	P430	P431	P432	P433	P434	P435	P436	P437	P438	P439	P440	P441	P442	P443	P444	P445	P446	P447	P448	P449	P450	P451	P452	P453	P454
Q393	Q394	Q395	Q396	Q397	Q398	Q399	Q400	Q401	Q402	Q403	Q404	Q405	Q406	Q407	Q408	Q409	Q410	Q411	Q412	Q413	Q414	Q415	Q416	Q417	Q418	Q419	Q420	Q421	Q422	Q423	Q424	Q425	Q426	Q427	Q428	Q429	Q430	Q431	Q432	Q433	Q434	Q435	Q436	Q437	Q438	Q439	Q440	Q441	Q442	Q443	Q444	Q445	Q446	Q447	Q448	Q449	Q450	Q451	Q452	Q453	Q454
R393	R394	R395	R396	R397	R398	R399	R400	R401	R402	R403	R404	R405	R406	R407	R408	R409	R410	R411	R412	R413	R414	R415	R416	R417	R418	R419	R420	R421	R422	R423	R424	R425	R426	R427	R428	R429	R430	R431	R432	R433	R434	R435	R436	R437	R438	R439	R440	R441	R442	R443	R444	R445	R446	R447	R448	R449	R450	R451	R452	R453	R454
S393	S394	S395	S396	S397	S398	S399	S400	S401	S402	S403	S404	S405	S406	S407	S408	S409	S410	S411	S412	S413	S414	S415	S416	S417	S418	S419	S420	S421	S422	S423	S424	S425	S426	S427	S428	S429	S430	S431	S432	S433	S434	S435	S436	S437	S438	S439	S440	S441	S442	S443	S444	S445	S446	S447	S448	S449	S450	S451	S452	S453	S454
T393	T394	T395	T396	T397	T398	T399	T400	T401	T402	T403	T404	T405	T406	T407	T408	T409	T410	T411	T412	T413	T414	T415	T416	T417	T418	T419	T420	T421	T422	T423	T424	T425	T426	T427	T428	T429	T430	T431	T432	T433	T434	T435	T436	T437	T438	T439	T440	T441	T442	T443	T444	T445	T446	T447	T448	T449	T450	T451	T452	T453	T454
U393	U394	U395	U396	U397	U398	U399	U400	U401	U402	U403	U404	U405	U406	U407	U408	U409	U410	U411	U412	U413	U414	U415	U416	U417	U418	U419	U420	U421	U422	U423	U424	U425	U426	U427	U428	U429	U430	U431	U432	U433	U434	U435	U436	U437	U438	U439	U440	U441	U442	U443	U444	U445	U446	U447	U448	U449	U450	U451	U452	U453	U454
V393	V394	V395	V396	V397	V398	V399	V400	V401	V402	V403	V404	V405	V406	V407	V408	V409	V410	V411	V412	V413	V414	V415	V416	V417	V418	V419	V420	V421	V422	V423	V424	V425	V426	V427	V428	V429	V430	V431	V432	V433	V434	V435	V436	V437	V438	V439	V440	V441	V442	V443	V444	V445	V446	V447	V448	V449	V450	V451	V452	V453	V454
W393	W394	W395	W396	W397	W398	W399	W400	W401	W402	W403	W404	W405	W406	W407	W408	W409	W410	W411	W412	W413	W414	W415	W416	W417	W418	W419	W420	W421	W422	W423	W424	W425	W426	W427	W428	W429	W430	W431	W432	W433	W434	W435	W436	W437	W438	W439	W440	W441	W442	W443	W444	W445	W446	W447	W448	W449	W450	W451	W452	W453	W454
X393	X394	X395	X396	X397	X398	X399	X400	X401	X402	X403	X404	X405	X406	X407	X408	X409	X410	X411	X412	X413	X414	X415	X416	X417	X418	X419	X420	X421	X422	X423	X424	X425	X426	X427	X428	X429	X430	X431	X432	X433	X434	X435	X436	X437	X438	X439	X440	X441	X442	X443	X444	X445	X446	X447	X448	X449	X450	X451	X452	X453	X454
Y393	Y394	Y395	Y396	Y397	Y398	Y399	Y400	Y401	Y402	Y403	Y404	Y405	Y406	Y407	Y408	Y409	Y410	Y411	Y412	Y413	Y414	Y415	Y416	Y417	Y418	Y419	Y420	Y421	Y422	Y423	Y424	Y425	Y426	Y427	Y428	Y429	Y430	Y431	Y432	Y433	Y434	Y435	Y436	Y437	Y438	Y439	Y440	Y441	Y442	Y443	Y444	Y445	Y446	Y447	Y448	Y449	Y450	Y451	Y452	Y453	Y454
Z393	Z394	Z395	Z396	Z397	Z398	Z399	Z400	Z401	Z402	Z403	Z404	Z405	Z406	Z407	Z408	Z409	Z410	Z411	Z412	Z413	Z414	Z415	Z416	Z417	Z418	Z419	Z420	Z421	Z422	Z423	Z424	Z425	Z426	Z427	Z428	Z429	Z430	Z431	Z432	Z433	Z434	Z435	Z436	Z437	Z438	Z439	Z440	Z441	Z442	Z443	Z444	Z445	Z446	Z447	Z448	Z449	Z450	Z451	Z452	Z453	Z454

5 Refinement protocol and experimental data overview

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

Of the 200 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
ARIA	refinement	2.3
TALOS	geometry optimization	3.70F1
CNS	structure solution	1.21
CNS	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	1435
Number of shifts mapped to atoms	1435
Number of unparsed shifts	0
Number of shifts with mapping errors	0
Number of shifts with mapping warnings	0
Assignment completeness (well-defined parts)	82%

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

COVALENT-GEOMETRY INFOmissingINFO

5.1 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	0	0	0	0±0
2	B	837	825	821	159±6
All	All	16740	16500	16420	3187

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 96.

5 of 338 unique clashes are listed below, sorted by their clash magnitude.

Atom-1	Atom-2	Clash(Å)	Distance(Å)	Models	
				Worst	Total
2:B:373:ALA:HB2	2:B:446:LEU:HD21	1.11	1.22	16	20
2:B:372:TYR:HA	2:B:442:MET:SD	0.92	2.03	17	20
2:B:372:TYR:CG	2:B:445:LYS:HD2	0.92	1.98	8	7
2:B:372:TYR:CD2	2:B:445:LYS:HG2	0.91	2.00	12	4
2:B:357:CYS:HB3	2:B:418:PHE:CD1	0.91	2.01	5	20

5.2 Torsion angles ⓘ

5.2.1 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	0	-	-	-	-	
2	B	102/128 (80%)	85±1 (83±1%)	15±1 (15±1%)	3±1 (3±1%)	9	45
All	All	2040/2820 (72%)	1691 (83%)	297 (15%)	52 (3%)	9	45

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)
2	B	387	LEU	20
2	B	390	TYR	12
2	B	414	ASP	12
2	B	373	ALA	7
2	B	367	LYS	1

5.2.2 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	0	-	-	-
2	B	89/113 (79%)	68±2 (77±2%)	21±2 (23±2%)	3 28
All	All	1780/2500 (71%)	1368 (77%)	412 (23%)	3 28

5 of 37 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)
2	B	403	ILE	20
2	B	368	LYS	20
2	B	361	LEU	20
2	B	365	PHE	20
2	B	392	ASP	20

5.2.3 RNA ⓘ

There are no RNA molecules in this entry.

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

1 non-standard protein/DNA/RNA residue is modelled in this entry.

In the following table, the Counts columns list the number of bonds for which Mogul statistics could be retrieved, the number of bonds that are observed in the model and the number of bonds that are defined in the chemical component dictionary. The Link column lists molecule types, if any, to which the group is linked. The Z score for a bond length is the number of standard deviations the observed value is removed from the expected value. A bond length with $|Z| > 2$ is considered an outlier worth inspection. RMSZ is the average root-mean-square of all Z scores of the bond lengths.

Mol	Type	Chain	Res	Link	Bond lengths		
					Counts	RMSZ	#Z>2
1	ALY	A	7	1	10,11,12	0.61±0.05	0±0 (0±0%)

In the following table, the Counts columns list the number of angles for which Mogul statistics could be retrieved, the number of angles that are observed in the model and the number of angles that are defined in the chemical component dictionary. The Link column lists molecule types, if any, to which the group is linked. The Z score for a bond angle is the number of standard deviations the observed value is removed from the expected value. A bond angle with $|Z| > 2$ is considered an outlier worth inspection. RMSZ is the average root-mean-square of all Z scores of the bond angles.

Mol	Type	Chain	Res	Link	Bond angles		
					Counts	RMSZ	#Z>2
1	ALY	A	7	1	7,12,14	0.91±0.02	0±0 (0±0%)

In the following table, the Chirals column lists the number of chiral outliers, the number of chiral centers analysed, the number of these observed in the model and the number defined in the chemical component dictionary. Similar counts are reported in the Torsion and Rings columns. '-' means no outliers of that kind were identified.

Mol	Type	Chain	Res	Link	Chirals	Torsions	Rings
1	ALY	A	7	1	-	0±0,9,10,12	-

There are no bond-length outliers.

There are no bond-angle outliers.

There are no chirality outliers.

There are no torsion outliers.

There are no ring outliers.

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 82% for the well-defined parts and 78% 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	1435
Number of shifts mapped to atoms	1435
Number of unparsed shifts	0
Number of shifts with mapping errors	0
Number of shifts with mapping warnings	0
Number of shift outliers (ShiftChecker)	19

6.1.2 Chemical shift referencing [i](#)

The following table shows the suggested chemical shift referencing corrections.

Nucleus	# values	Correction \pm precision, ppm	Suggested action
$^{13}\text{C}_\alpha$	127	-0.45 ± 0.27	None needed (< 0.5 ppm)
$^{13}\text{C}_\beta$	123	0.32 ± 0.17	None needed (< 0.5 ppm)
$^{13}\text{C}'$	10	—	None (insufficient data)
^{15}N	114	0.41 ± 0.29	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 82%, i.e. 1084 atoms were assigned a chemical shift out of a possible 1318. 13 out of 13 assigned methyl groups (LEU and VAL) were assigned stereospecifically.

	Total	^1H	^{13}C	^{15}N
Backbone	397/500 (79%)	192/199 (96%)	110/204 (54%)	95/97 (98%)
Sidechain	582/672 (87%)	367/400 (92%)	213/242 (88%)	2/30 (7%)

Continued on next page...

Continued from previous page...

	Total	¹H	¹³C	¹⁵N
Aromatic	105/146 (72%)	68/76 (89%)	36/61 (59%)	1/9 (11%)
Overall	1084/1318 (82%)	627/675 (93%)	359/507 (71%)	98/136 (72%)

6.1.4 Statistically unusual chemical shifts ⓘ

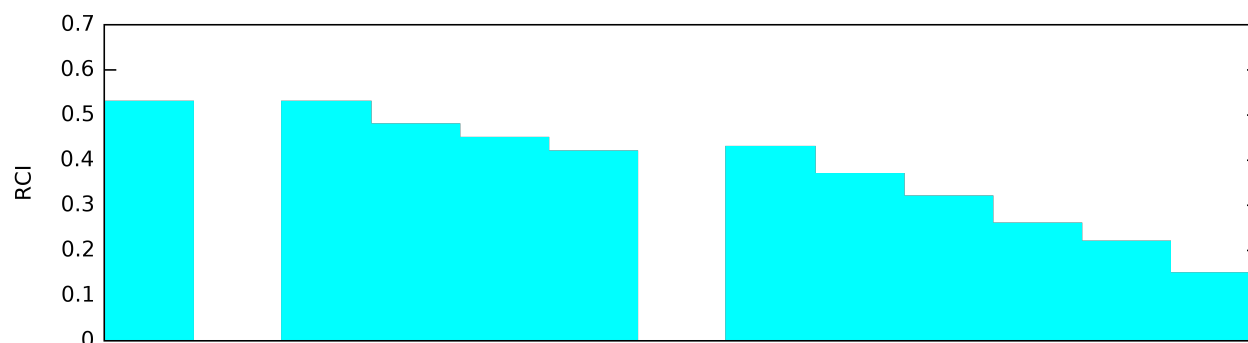
The following table lists the statistically unusual chemical shifts. These are statistical measures, and large deviations from the mean do not necessarily imply incorrect assignments. Molecules containing paramagnetic centres or hemes are expected to give rise to anomalous chemical shifts.

Mol	Chain	Res	Type	Atom	Shift, ppm	Expected range, ppm	Z-score
2	B	347	SER	C	8.45	183.48 – 165.88	-94.4
2	B	386	GLY	C	7.90	183.33 – 164.53	-88.3
2	B	346	LYS	C	8.51	186.58 – 166.78	-84.9
2	B	385	LEU	C	7.48	187.06 – 166.96	-84.3
2	B	430	TYR	C	7.61	185.42 – 165.42	-83.9
2	B	447	GLN	C	11.57	186.20 – 166.50	-83.6
2	B	388	HIS	C	8.30	185.27 – 165.27	-83.5
2	B	437	HIS	C	9.41	185.27 – 165.27	-82.9
2	B	384	ALA	C	7.86	188.57 – 166.97	-78.7
2	B	366	ALA	C	9.11	188.57 – 166.97	-78.1
2	B	375	PRO	HG2	-0.72	3.48 – 0.38	-8.6
2	B	447	GLN	HB2	0.07	3.30 – 0.80	-7.9
2	B	375	PRO	HB2	-0.35	3.82 – 0.32	-6.9
2	B	357	CYS	HB2	0.35	5.20 – 0.70	-5.8
2	B	422	VAL	HG11	-0.67	2.13 – -0.47	-5.8
2	B	422	VAL	HG13	-0.67	2.13 – -0.47	-5.8
2	B	422	VAL	HG12	-0.67	2.13 – -0.47	-5.8
2	B	375	PRO	HD2	1.62	5.45 – 1.85	-5.6
2	B	439	VAL	H	11.70	11.69 – 4.89	5.0

6.1.5 Random Coil Index (RCI) plots ⓘ

The images below report *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:



Random coil index (RCI) for chain B:

