Full wwPDB X-ray Structure Validation Report ⓘ

Sep 1, 2022 – 04:20 pm BST

PDB ID	:	6G5K
Title	:	Crystal structure of the binding domain of Botulinum Neurotoxin type B in complex with human synaptotagmin 1
Authors	:	Masuyer, G.; Elliot, M.; Favre-Guilmard, C.; Liu, S.M.; Maignel, J.; Beard, M.; Carre, D.; Kalinichev, M.; Lezmi, S.; Mir, I.; Nicoleau, C.; Palan, S.; Perier, C.; Raban, E.; Dong, M.; Krupp, J.; Stenmark, P.
Deposited on	:	2018-03-29
Resolution	:	2.00 Å(reported)

This is a Full wwPDB X-ray 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/XrayValidationReportHelp
with specific help available everywhere you see the ⓘ 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 ⓘ) were used in the production of this report:

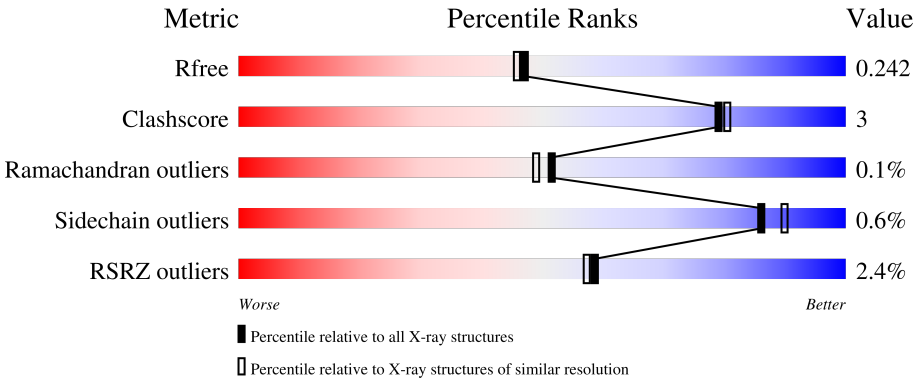
MolProbity	:	4.02b-467
Xtriage (Phenix)	:	1.13
EDS	:	2.30
Percentile statistics	:	20191225.v01 (using entries in the PDB archive December 25th 2019)
Refmac	:	5.8.0267
CCP4	:	7.1.010 (Gargrove)
Ideal geometry (proteins)	:	Engh & Huber (2001)
Ideal geometry (DNA, RNA)	:	Parkinson et al. (1996)
Validation Pipeline (wwPDB-VP)	:	2.30

1 Overall quality at a glance (i)

The following experimental techniques were used to determine the structure:
X-RAY DIFFRACTION

The reported resolution of this entry is 2.00 Å.

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)	Similar resolution (#Entries, resolution range(Å))
R_{free}	130704	8085 (2.00-2.00)
Clashscore	141614	9178 (2.00-2.00)
Ramachandran outliers	138981	9054 (2.00-2.00)
Sidechain outliers	138945	9053 (2.00-2.00)
RSRZ outliers	127900	7900 (2.00-2.00)

The table below summarises the geometric issues observed across the polymeric chains and their fit to the electron density. The red, orange, yellow and green segments of the lower bar indicate the fraction of residues that contain outliers for >=3, 2, 1 and 0 types of geometric quality criteria respectively. 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% The upper red bar (where present) indicates the fraction of residues that have poor fit to the electron density. The numeric value is given above the bar.

Mol	Chain	Length	Quality of chain
1	A	461	2% 85% 5% 9%
1	B	461	2% 84% 8% 8%
2	33	21	5% 57% 14% 29%
2	C	21	14% 67% 10% 24%

2 Entry composition (i)

There are 3 unique types of molecules in this entry. The entry contains 7923 atoms, of which 0 are hydrogens and 0 are deuteriums.

In the tables below, the ZeroOcc column contains the number of atoms modelled with zero occupancy, the AltConf column contains the number of residues with at least one atom in alternate conformation and the Trace column contains the number of residues modelled with at most 2 atoms.

- Molecule 1 is a protein called Botulinum neurotoxin type B.

Mol	Chain	Residues	Atoms					ZeroOcc	AltConf	Trace
1	A	421	Total	C	N	O	S	0	1	0
			3595	2325	590	672	8			
1	B	424	Total	C	N	O	S	0	2	0
			3624	2343	597	676	8			

There are 52 discrepancies between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	Comment	Reference
A	833	MET	-	initiating methionine	UNP P10844
A	834	GLY	-	expression tag	UNP P10844
A	835	SER	-	expression tag	UNP P10844
A	836	SER	-	expression tag	UNP P10844
A	837	HIS	-	expression tag	UNP P10844
A	838	HIS	-	expression tag	UNP P10844
A	839	HIS	-	expression tag	UNP P10844
A	840	HIS	-	expression tag	UNP P10844
A	841	HIS	-	expression tag	UNP P10844
A	842	HIS	-	expression tag	UNP P10844
A	843	SER	-	expression tag	UNP P10844
A	844	SER	-	expression tag	UNP P10844
A	845	GLY	-	expression tag	UNP P10844
A	846	LEU	-	expression tag	UNP P10844
A	847	VAL	-	expression tag	UNP P10844
A	848	PRO	-	expression tag	UNP P10844
A	849	ARG	-	expression tag	UNP P10844
A	850	GLY	-	expression tag	UNP P10844
A	851	SER	-	expression tag	UNP P10844
A	852	HIS	-	expression tag	UNP P10844
A	853	MET	-	expression tag	UNP P10844
A	854	ALA	-	expression tag	UNP P10844
A	855	SER	-	expression tag	UNP P10844
A	856	MET	-	expression tag	UNP P10844
A	1292	LEU	-	expression tag	UNP P10844

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Chain	Residue	Modelled	Actual	Comment	Reference
A	1293	GLN	-	expression tag	UNP P10844
B	833	MET	-	initiating methionine	UNP P10844
B	834	GLY	-	expression tag	UNP P10844
B	835	SER	-	expression tag	UNP P10844
B	836	SER	-	expression tag	UNP P10844
B	837	HIS	-	expression tag	UNP P10844
B	838	HIS	-	expression tag	UNP P10844
B	839	HIS	-	expression tag	UNP P10844
B	840	HIS	-	expression tag	UNP P10844
B	841	HIS	-	expression tag	UNP P10844
B	842	HIS	-	expression tag	UNP P10844
B	843	SER	-	expression tag	UNP P10844
B	844	SER	-	expression tag	UNP P10844
B	845	GLY	-	expression tag	UNP P10844
B	846	LEU	-	expression tag	UNP P10844
B	847	VAL	-	expression tag	UNP P10844
B	848	PRO	-	expression tag	UNP P10844
B	849	ARG	-	expression tag	UNP P10844
B	850	GLY	-	expression tag	UNP P10844
B	851	SER	-	expression tag	UNP P10844
B	852	HIS	-	expression tag	UNP P10844
B	853	MET	-	expression tag	UNP P10844
B	854	ALA	-	expression tag	UNP P10844
B	855	SER	-	expression tag	UNP P10844
B	856	MET	-	expression tag	UNP P10844
B	1292	LEU	-	expression tag	UNP P10844
B	1293	GLN	-	expression tag	UNP P10844

- Molecule 2 is a protein called Synaptotagmin-1.

Mol	Chain	Residues	Atoms					ZeroOcc	AltConf	Trace
2	C	16	Total	C	N	O	S	0	0	0
			137	89	23	24	1			
2	33	15	Total	C	N	O	S	0	0	0
			128	83	21	23	1			

- Molecule 3 is water.

Mol	Chain	Residues	Atoms		ZeroOcc	AltConf
3	A	212	Total	O	0	0
			212	212		

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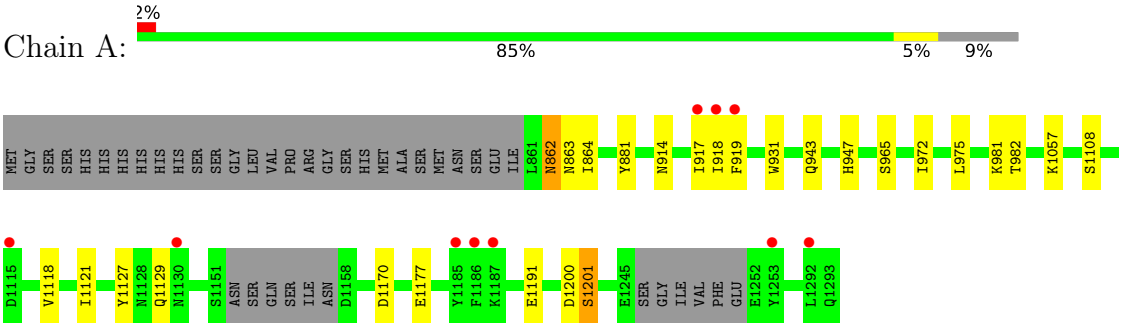
Mol	Chain	Residues	Atoms	ZeroOcc	AltConf
3	C	3	Total O 3 3	0	0
3	B	219	Total O 219 219	0	0
3	33	5	Total O 5 5	0	0

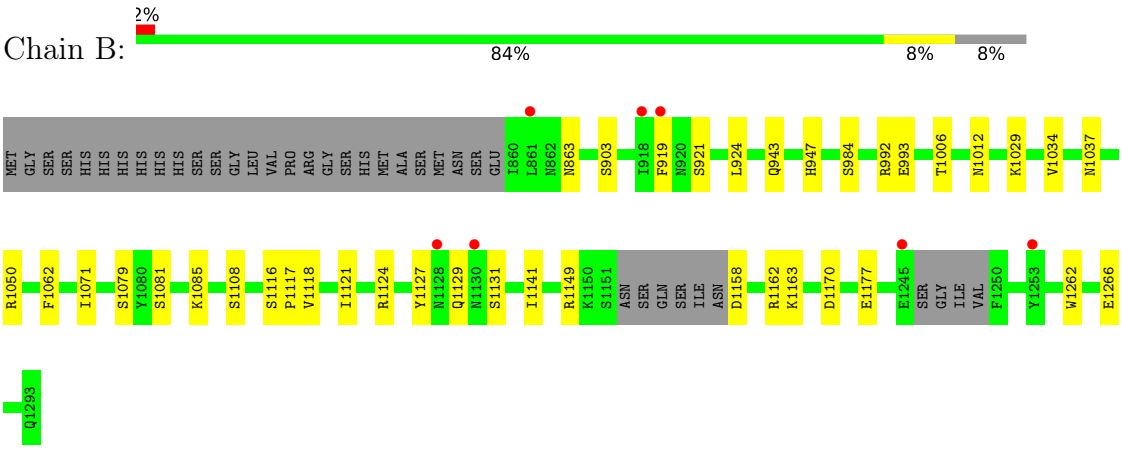
3 Residue-property plots (i)

These plots are drawn for all protein, RNA, DNA and oligosaccharide chains in the entry. The first graphic for a chain summarises the proportions of the various outlier classes displayed in the second graphic. The second graphic shows the sequence view annotated by issues in geometry and electron density. Residues are color-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. A red dot above a residue indicates a poor fit to the electron density (RSRZ > 2). Stretches of 2 or more consecutive residues without any outlier are shown as a green connector. Residues present in the sample, but not in the model, are shown in grey.

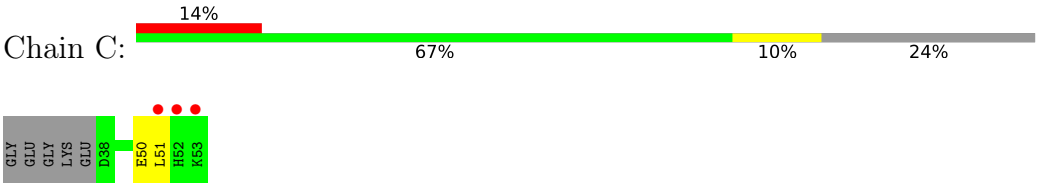
• Molecule 1: Botulinum neurotoxin type B



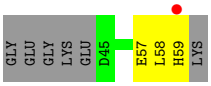
• Molecule 1: Botulinum neurotoxin type B



• Molecule 2: Synaptotagmin-1



• Molecule 2: Synaptotagmin-1


GLY
GLU
GLY
LYS
GLU
D45
E57
L58
H59
LYS

4 Data and refinement statistics (i)

Property	Value	Source
Space group	P 1 21 1	Depositor
Cell constants a, b, c, α, β, γ	48.30Å 212.79Å 52.91Å 90.00° 91.08° 90.00°	Depositor
Resolution (Å)	53.25 – 2.00 53.20 – 2.00	Depositor EDS
% Data completeness (in resolution range)	99.9 (53.25-2.00) 99.9 (53.20-2.00)	Depositor EDS
R_{merge}	0.10	Depositor
R_{sym}	(Not available)	Depositor
$< I/\sigma(I) >$ [1]	1.58 (at 2.00Å)	Xtriage
Refinement program	REFMAC 5.8.0222	Depositor
R, R_{free}	0.194 , 0.235 0.201 , 0.242	Depositor DCC
R_{free} test set	3555 reflections (4.96%)	wwPDB-VP
Wilson B-factor (Å^2)	35.1	Xtriage
Anisotropy	0.620	Xtriage
Bulk solvent k_{sol}(e/Å^3), B_{sol}(Å^2)	(Not available) , (Not available)	EDS
L-test for twinning[2]	$< \|L\| > = 0.51$, $< L^2 > = 0.35$	Xtriage
Estimated twinning fraction	0.277 for h,-k,-l	Xtriage
F_o,F_c correlation	0.96	EDS
Total number of atoms	7923	wwPDB-VP
Average B, all atoms (Å^2)	45.0	wwPDB-VP

Xtriage's analysis on translational NCS is as follows: *The largest off-origin peak in the Patterson function is 5.53% of the height of the origin peak. No significant pseudotranslation is detected.*

[1] Intensities estimated from amplitudes.

[2] Theoretical values of $< |L| >$, $< L^2 >$ for acentric reflections are 0.5, 0.333 respectively for untwinned datasets, and 0.375, 0.2 for perfectly twinned datasets.

5 Model quality

5.1 Standard geometry

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 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	0.40	0/3679	0.62	0/4958
1	B	0.41	0/3708	0.62	0/4997
2	33	0.47	0/130	0.53	0/170
2	C	0.42	0/139	0.55	0/181
All	All	0.41	0/7656	0.62	0/10306

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 outliers	#Planarity outliers
1	B	0	2

There are no bond length outliers.

There are no bond angle outliers.

There are no chirality outliers.

All (2) planarity outliers are listed below:

Mol	Chain	Res	Type	Group
1	B	1162	ARG	Sidechain
1	B	992	ARG	Sidechain

5.2 Too-close contacts

In the following table, the Non-H and H(model) columns list the number of non-hydrogen atoms and hydrogen atoms in the 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 within the asymmetric unit, whereas Symm-Clashes lists symmetry-related clashes.

Mol	Chain	Non-H	H(model)	H(added)	Clashes	Symm-Clashes
1	A	3595	0	3510	16	0
1	B	3624	0	3537	24	0
2	33	128	0	126	2	0
2	C	137	0	139	2	0
3	33	5	0	0	0	0
3	A	212	0	0	1	0
3	B	219	0	0	1	0
3	C	3	0	0	0	0
All	All	7923	0	7312	41	0

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 3.

All (41) close contacts within the same asymmetric unit are listed below, sorted by their clash magnitude.

Atom-1	Atom-2	Interatomic distance (Å)	Clash overlap (Å)
1:B:919:PHE:CZ	1:B:1037:ASN:HB3	1.97	0.99
1:B:919:PHE:HZ	1:B:1037:ASN:HB3	1.37	0.84
1:A:914:ASN:O	1:A:918:ILE:HG22	1.80	0.81
1:B:919:PHE:HZ	1:B:1037:ASN:CB	1.94	0.80
1:B:919:PHE:HZ	1:B:1037:ASN:CA	1.99	0.74
1:B:924:LEU:HD21	1:B:1034:VAL:HB	1.79	0.64
1:B:903:SER:OG	1:B:1050:ARG:NH1	2.32	0.62
1:A:1127:TYR:CZ	1:A:1129:GLN:HB2	2.36	0.60
2:33:58:LEU:O	2:33:59:HIS:CG	2.56	0.58
1:B:1118:VAL:HG11	2:33:57:GLU:HG2	1.87	0.57
1:A:1129:GLN:N	1:A:1129:GLN:OE1	2.39	0.56
1:B:919:PHE:CZ	1:B:1037:ASN:CB	2.75	0.53
1:A:1118:VAL:HG11	2:C:50:GLU:HG2	1.90	0.52
1:B:1127:TYR:OH	1:B:1129:GLN:HB2	2.09	0.51
1:B:1127:TYR:CZ	1:B:1129:GLN:HB2	2.47	0.50
1:B:1116:SER:HB2	1:B:1117:PRO:HD2	1.94	0.49
1:B:943:GLN:O	1:B:947:HIS:HD2	1.95	0.49
1:B:1079:SER:O	1:B:1085:LYS:NZ	2.47	0.47
1:A:975:LEU:O	1:A:982:THR:HA	2.15	0.46
1:B:919:PHE:CE2	1:B:1037:ASN:HB3	2.46	0.46
1:B:1081:SER:O	1:B:1163:LYS:NZ	2.49	0.46
1:B:993:GLU:HB2	3:B:1394:HOH:O	2.15	0.46
1:A:862:ASN:O	1:A:863:ASN:HB2	2.16	0.45
1:B:1262:TRP:CH2	1:B:1266:GLU:HG3	2.52	0.44
1:B:1149:ARG:NH2	1:B:1158:ASP:O	2.48	0.44
1:A:1108:SER:HB3	1:A:1121:ILE:CG2	2.47	0.43

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Atom-1	Atom-2	Interatomic distance (Å)	Clash overlap (Å)
1:A:965:SER:HB3	1:A:972:ILE:HB	2.00	0.43
1:B:1108:SER:HB3	1:B:1121:ILE:CG2	2.49	0.42
1:A:881:TYR:CE1	1:A:917:ILE:HG21	2.54	0.42
1:A:931:TRP:HB2	1:A:1057:LYS:HG2	2.01	0.42
1:A:1200:ASP:O	1:A:1201:SER:C	2.58	0.42
1:A:1191:GLU:CD	2:C:51:LEU:CD1	2.88	0.41
1:B:863:ASN:O	1:B:1062:PHE:HA	2.21	0.41
1:A:864:ILE:O	1:A:864:ILE:HG23	2.20	0.41
1:A:1170:ASP:OD2	1:A:1177:GLU:OE2	2.38	0.41
1:B:1170:ASP:OD2	1:B:1177:GLU:OE2	2.38	0.41
1:B:1006:THR:HG21	1:B:1071:ILE:HG12	2.03	0.41
1:A:919:PHE:HB3	3:A:1477:HOH:O	2.21	0.41
1:B:1012:ASN:OD1	1:B:1029:LYS:HG2	2.20	0.41
1:B:1124:ARG:HG2	1:B:1141:ILE:HD11	2.03	0.41
1:A:943:GLN:O	1:A:947:HIS:HD2	2.05	0.40

There are no symmetry-related clashes.

5.3 Torsion angles (i)

5.3.1 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 X-ray entries followed by that with respect to entries of similar resolution.

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	416/461 (90%)	397 (95%)	18 (4%)	1 (0%)	47	44
1	B	420/461 (91%)	399 (95%)	21 (5%)	0	100	100
2	33	13/21 (62%)	12 (92%)	1 (8%)	0	100	100
2	C	14/21 (67%)	13 (93%)	1 (7%)	0	100	100
All	All	863/964 (90%)	821 (95%)	41 (5%)	1 (0%)	51	49

All (1) Ramachandran outliers are listed below:

Mol	Chain	Res	Type
1	A	1201	SER

5.3.2 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 X-ray entries followed by that with respect to entries of similar resolution.

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	396/432 (92%)	394 (100%)	2 (0%)	88 92
1	B	398/432 (92%)	395 (99%)	3 (1%)	81 86
2	33	14/18 (78%)	14 (100%)	0	100 100
2	C	15/18 (83%)	15 (100%)	0	100 100
All	All	823/900 (91%)	818 (99%)	5 (1%)	86 90

All (5) residues with a non-rotameric sidechain are listed below:

Mol	Chain	Res	Type
1	A	862	ASN
1	A	981	LYS
1	B	921	SER
1	B	984	SER
1	B	1131	SER

Sometimes sidechains can be flipped to improve hydrogen bonding and reduce clashes. All (5) such sidechains are listed below:

Mol	Chain	Res	Type
1	A	914	ASN
1	A	947	HIS
1	B	947	HIS
1	B	1052	GLN
1	B	1128	ASN

5.3.3 RNA (i)

There are no RNA molecules in this entry.

5.4 Non-standard residues in protein, DNA, RNA chains (i)

There are no non-standard protein/DNA/RNA residues in this entry.

5.5 Carbohydrates (i)

There are no monosaccharides in this entry.

5.6 Ligand geometry (i)

There are no ligands in this entry.

5.7 Other polymers (i)

There are no such residues in this entry.

5.8 Polymer linkage issues (i)

There are no chain breaks in this entry.

6 Fit of model and data

6.1 Protein, DNA and RNA chains

In the following table, the column labelled '#RSRZ> 2' contains the number (and percentage) of RSRZ outliers, followed by percent RSRZ outliers for the chain as percentile scores relative to all X-ray entries and entries of similar resolution. The OWAB column contains the minimum, median, 95^{th} percentile and maximum values of the occupancy-weighted average B-factor per residue. The column labelled 'Q< 0.9' lists the number of (and percentage) of residues with an average occupancy less than 0.9.

Mol	Chain	Analysed	<RSRZ>	#RSRZ>2	OWAB(Å^2)	Q<0.9
1	A	421/461 (91%)	-0.26	10 (2%) 59 57	29, 41, 79, 118	0
1	B	424/461 (91%)	-0.25	7 (1%) 70 68	26, 39, 78, 155	0
2	33	15/21 (71%)	0.18	1 (6%) 17 17	45, 55, 76, 86	0
2	C	16/21 (76%)	0.46	3 (18%) 1 1	46, 57, 93, 97	0
All	All	876/964 (90%)	-0.24	21 (2%) 59 57	26, 41, 81, 155	0

All (21) RSRZ outliers are listed below:

Mol	Chain	Res	Type	RSRZ
1	B	919	PHE	12.6
1	B	1130	ASN	7.0
1	A	1130	ASN	6.2
1	B	918	ILE	5.0
1	A	918	ILE	4.3
1	A	919	PHE	3.9
2	33	59	HIS	3.8
2	C	52	HIS	3.6
2	C	51	LEU	3.4
1	B	861	LEU	3.0
1	A	917	ILE	2.8
1	A	1185	TYR	2.6
1	B	1253	TYR	2.6
1	A	1186	PHE	2.6
1	A	1187	LYS	2.5
1	A	1292	LEU	2.4
2	C	53	LYS	2.3
1	A	1115	ASP	2.3
1	A	1253	TYR	2.2
1	B	1245	GLU	2.1
1	B	1128	ASN	2.0

6.2 Non-standard residues in protein, DNA, RNA chains (i)

There are no non-standard protein/DNA/RNA residues in this entry.

6.3 Carbohydrates (i)

There are no monosaccharides in this entry.

6.4 Ligands (i)

There are no ligands in this entry.

6.5 Other polymers (i)

There are no such residues in this entry.