



Full wwPDB X-ray Structure Validation Report ⓘ

Apr 7, 2025 – 01:04 pm BST

PDB ID : 9EWR / pdb_00009ewr
Title : Crystal structure of an inverse charged cutinase mutant from *Saccharopolyspora flava* (611)
Authors : Zahn, M.; Green, K.; Oliveira, L.; Lichtenstein, B.R.
Deposited on : 2024-04-04
Resolution : 1.17 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	:	3.0
Percentile statistics	:	20231227.v01 (using entries in the PDB archive December 27th 2023)
CCP4	:	9.0.003 (Gargrove)
Density-Fitness	:	1.0.11
Ideal geometry (proteins)	:	Engh & Huber (2001)
Ideal geometry (DNA, RNA)	:	Parkinson et al. (1996)
Validation Pipeline (wwPDB-VP)	:	2.42

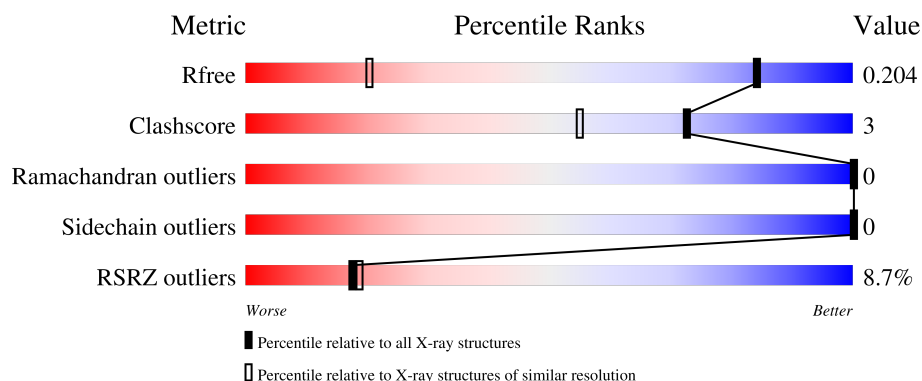
1 Overall quality at a glance

The following experimental techniques were used to determine the structure:

X-RAY DIFFRACTION

The reported resolution of this entry is 1.17 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}	164625	1569 (1.20-1.16)
Clashscore	180529	1711 (1.20-1.16)
Ramachandran outliers	177936	1657 (1.20-1.16)
Sidechain outliers	177891	1657 (1.20-1.16)
RSRZ outliers	164620	1568 (1.20-1.16)

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 $\leq 5\%$. 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	269	<div> <div>5%</div> <div>88%</div> <div>6% . .</div> </div>
1	B	269	<div> <div>12%</div> <div>90%</div> <div>6% .</div> </div>

2 Entry composition

There are 2 unique types of molecules in this entry. The entry contains 4493 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 cutinase.

Mol	Chain	Residues	Atoms					ZeroOcc	AltConf	Trace
1	A	257	Total 2013	C 1279	N 354	O 375	S 5	0	5	0
1	B	259	Total 2018	C 1280	N 358	O 375	S 5	0	2	0

There are 64 discrepancies between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	Comment	Reference
A	1	MET	-	initiating methionine	UNP A0A1I6NU60
A	6	LYS	ASP	engineered mutation	UNP A0A1I6NU60
A	11	LYS	ASP	engineered mutation	UNP A0A1I6NU60
A	19	LYS	ALA	engineered mutation	UNP A0A1I6NU60
A	27	ASN	ASP	engineered mutation	UNP A0A1I6NU60
A	28	ARG	GLU	engineered mutation	UNP A0A1I6NU60
A	29	ARG	GLU	engineered mutation	UNP A0A1I6NU60
A	35	LYS	SER	engineered mutation	UNP A0A1I6NU60
A	37	LYS	SER	engineered mutation	UNP A0A1I6NU60
A	49	ASN	ASP	engineered mutation	UNP A0A1I6NU60
A	109	LYS	ASP	engineered mutation	UNP A0A1I6NU60
A	114	LYS	ASP	engineered mutation	UNP A0A1I6NU60
A	116	LYS	ASP	engineered mutation	UNP A0A1I6NU60
A	147	LYS	ALA	engineered mutation	UNP A0A1I6NU60
A	164	ARG	SER	engineered mutation	UNP A0A1I6NU60
A	167	ARG	GLN	engineered mutation	UNP A0A1I6NU60
A	188	GLU	LYS	engineered mutation	UNP A0A1I6NU60
A	192	LYS	GLU	engineered mutation	UNP A0A1I6NU60
A	197	LYS	ASP	engineered mutation	UNP A0A1I6NU60
A	240	ARG	GLN	engineered mutation	UNP A0A1I6NU60
A	247	ARG	GLU	engineered mutation	UNP A0A1I6NU60
A	249	ASN	ASP	engineered mutation	UNP A0A1I6NU60
A	250	LYS	ASP	engineered mutation	UNP A0A1I6NU60
A	256	ARG	GLN	engineered mutation	UNP A0A1I6NU60
A	262	LYS	-	expression tag	UNP A0A1I6NU60

Continued on next page...

Continued from previous page...

Chain	Residue	Modelled	Actual	Comment	Reference
A	263	GLU	-	expression tag	UNP A0A1I6NU60
A	264	HIS	-	expression tag	UNP A0A1I6NU60
A	265	HIS	-	expression tag	UNP A0A1I6NU60
A	266	HIS	-	expression tag	UNP A0A1I6NU60
A	267	HIS	-	expression tag	UNP A0A1I6NU60
A	268	HIS	-	expression tag	UNP A0A1I6NU60
A	269	HIS	-	expression tag	UNP A0A1I6NU60
B	1	MET	-	initiating methionine	UNP A0A1I6NU60
B	6	LYS	ASP	engineered mutation	UNP A0A1I6NU60
B	11	LYS	ASP	engineered mutation	UNP A0A1I6NU60
B	19	LYS	ALA	engineered mutation	UNP A0A1I6NU60
B	27	ASN	ASP	engineered mutation	UNP A0A1I6NU60
B	28	ARG	GLU	engineered mutation	UNP A0A1I6NU60
B	29	ARG	GLU	engineered mutation	UNP A0A1I6NU60
B	35	LYS	SER	engineered mutation	UNP A0A1I6NU60
B	37	LYS	SER	engineered mutation	UNP A0A1I6NU60
B	49	ASN	ASP	engineered mutation	UNP A0A1I6NU60
B	109	LYS	ASP	engineered mutation	UNP A0A1I6NU60
B	114	LYS	ASP	engineered mutation	UNP A0A1I6NU60
B	116	LYS	ASP	engineered mutation	UNP A0A1I6NU60
B	147	LYS	ALA	engineered mutation	UNP A0A1I6NU60
B	164	ARG	SER	engineered mutation	UNP A0A1I6NU60
B	167	ARG	GLN	engineered mutation	UNP A0A1I6NU60
B	188	GLU	LYS	engineered mutation	UNP A0A1I6NU60
B	192	LYS	GLU	engineered mutation	UNP A0A1I6NU60
B	197	LYS	ASP	engineered mutation	UNP A0A1I6NU60
B	240	ARG	GLN	engineered mutation	UNP A0A1I6NU60
B	247	ARG	GLU	engineered mutation	UNP A0A1I6NU60
B	249	ASN	ASP	engineered mutation	UNP A0A1I6NU60
B	250	LYS	ASP	engineered mutation	UNP A0A1I6NU60
B	256	ARG	GLN	engineered mutation	UNP A0A1I6NU60
B	262	LYS	-	expression tag	UNP A0A1I6NU60
B	263	GLU	-	expression tag	UNP A0A1I6NU60
B	264	HIS	-	expression tag	UNP A0A1I6NU60
B	265	HIS	-	expression tag	UNP A0A1I6NU60
B	266	HIS	-	expression tag	UNP A0A1I6NU60
B	267	HIS	-	expression tag	UNP A0A1I6NU60
B	268	HIS	-	expression tag	UNP A0A1I6NU60
B	269	HIS	-	expression tag	UNP A0A1I6NU60

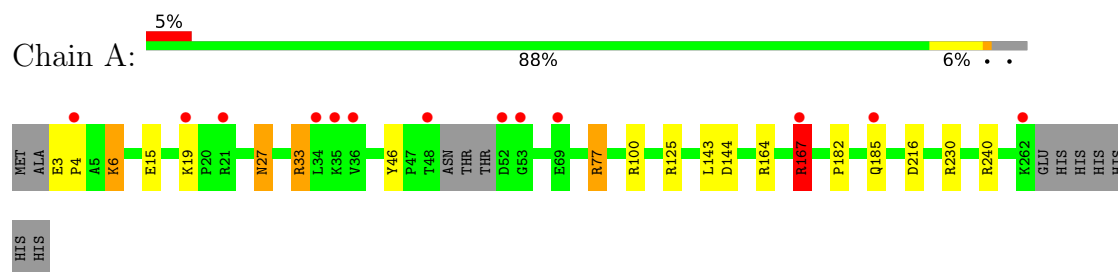
- Molecule 2 is water.

Mol	Chain	Residues	Atoms		ZeroOcc	AltConf
2	A	249	Total 249	O 249	0	0
2	B	213	Total 213	O 213	0	0

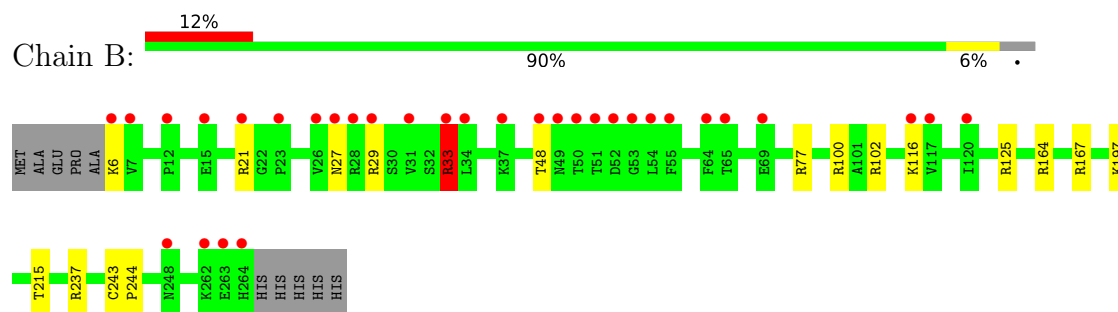
3 Residue-property plots [i](#)

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: cutinase



- Molecule 1: cutinase



4 Data and refinement statistics

Property	Value	Source
Space group	P 1 21 1	Depositor
Cell constants a, b, c, α , β , γ	46.08Å 41.32Å 124.09Å 90.00° 99.81° 90.00°	Depositor
Resolution (Å)	122.27 – 1.17 122.27 – 1.17	Depositor EDS
% Data completeness (in resolution range)	63.7 (122.27-1.17) 63.7 (122.27-1.17)	Depositor EDS
R_{merge}	0.08	Depositor
R_{sym}	(Not available)	Depositor
$\langle I/\sigma(I) \rangle$ ¹	1.88 (at 1.17Å)	Xtriage
Refinement program	REFMAC 5.8.0425	Depositor
R, R_{free}	0.186 , 0.204 0.186 , 0.204	Depositor DCC
R_{free} test set	4902 reflections (4.95%)	wwPDB-VP
Wilson B-factor (Å ²)	11.2	Xtriage
Anisotropy	0.079	Xtriage
Bulk solvent k_{sol} (e/Å ³), B_{sol} (Å ²)	0.39 , 36.1	EDS
L-test for twinning ²	$\langle L \rangle = 0.48$, $\langle L^2 \rangle = 0.31$	Xtriage
Estimated twinning fraction	0.027 for h,-k,-h-l	Xtriage
F_o, F_c correlation	0.95	EDS
Total number of atoms	4493	wwPDB-VP
Average B, all atoms (Å ²)	15.0	wwPDB-VP

Xtriage's analysis on translational NCS is as follows: *The largest off-origin peak in the Patterson function is 10.20% of the height of the origin peak. No significant pseudotranslation is detected.*

¹Intensities estimated from amplitudes.

²Theoretical values of $\langle |L| \rangle$, $\langle L^2 \rangle$ for acentric reflections are 0.5, 0.333 respectively for untwinned datasets, and 0.375, 0.2 for perfectly twinned datasets.

5 Model quality [i](#)

5.1 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 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	0.52	0/2080	1.07	12/2824 (0.4%)
1	B	0.52	0/2077	0.97	4/2821 (0.1%)
All	All	0.52	0/4157	1.02	16/5645 (0.3%)

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	A	0	5
1	B	0	7
All	All	0	12

There are no bond length outliers.

All (16) bond angle outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(°)	Ideal(°)
1	A	230	ARG	NE-CZ-NH1	8.08	124.34	120.30
1	A	167	ARG	NE-CZ-NH2	-7.85	116.38	120.30
1	A	33[A]	ARG	CG-CD-NE	-7.55	95.95	111.80
1	A	33[B]	ARG	CG-CD-NE	-7.55	95.95	111.80
1	B	33	ARG	CG-CD-NE	-7.07	96.95	111.80
1	B	33	ARG	NE-CZ-NH2	-6.84	116.88	120.30
1	A	230	ARG	NE-CZ-NH2	-6.16	117.22	120.30
1	B	77	ARG	NE-CZ-NH2	-5.96	117.32	120.30
1	A	143	LEU	CB-CG-CD2	-5.82	101.11	111.00
1	A	6	LYS	CB-CG-CD	5.61	126.18	111.60
1	A	27[A]	ASN	N-CA-CB	5.53	120.55	110.60
1	A	27[B]	ASN	N-CA-CB	5.53	120.55	110.60
1	A	33[A]	ARG	N-CA-CB	-5.27	101.11	110.60
1	A	33[B]	ARG	N-CA-CB	-5.27	101.11	110.60

Continued on next page...

Continued from previous page...

Mol	Chain	Res	Type	Atoms	Z	Observed(°)	Ideal(°)
1	A	77	ARG	NE-CZ-NH2	-5.11	117.75	120.30
1	B	102	ARG	NE-CZ-NH1	5.07	122.83	120.30

There are no chirality outliers.

All (12) planarity outliers are listed below:

Mol	Chain	Res	Type	Group
1	A	100	ARG	Sidechain
1	A	164	ARG	Sidechain
1	A	167	ARG	Sidechain
1	A	33[A]	ARG	Sidechain
1	A	33[B]	ARG	Sidechain
1	B	100	ARG	Sidechain
1	B	164	ARG	Sidechain
1	B	167	ARG	Sidechain
1	B	21[A]	ARG	Sidechain
1	B	21[B]	ARG	Sidechain
1	B	237	ARG	Sidechain
1	B	33	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	2013	0	2024	13	0
1	B	2018	0	2026	12	0
2	A	249	0	0	3	1
2	B	213	0	0	2	3
All	All	4493	0	4050	25	3

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 (25) 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:A:77:ARG:NH1	1:A:216[A]:ASP:OD1	2.11	0.83
1:A:167:ARG:HD3	2:A:356:HOH:O	1.89	0.71
1:A:144:ASP:OD1	2:A:301:HOH:O	2.09	0.70
1:B:215:THR:HG23	2:B:331:HOH:O	1.97	0.63
1:A:77:ARG:NH1	1:A:216[A]:ASP:CG	2.53	0.62
1:B:6:LYS:CE	1:B:125:ARG:HH21	2.13	0.60
1:A:240:ARG:NH2	2:A:313:HOH:O	2.39	0.55
1:A:3:GLU:N	1:A:4:PRO:HD2	2.24	0.53
1:B:6:LYS:HE2	1:B:125:ARG:HH21	1.74	0.52
1:A:77:ARG:NH1	1:A:216[A]:ASP:OD2	2.42	0.52
1:B:6:LYS:HE2	1:B:125:ARG:NH2	2.26	0.51
1:B:27:ASN:OD1	1:B:48:THR:HG22	2.11	0.50
1:A:3:GLU:N	1:A:4:PRO:CD	2.74	0.49
1:A:77:ARG:HH11	1:A:216[A]:ASP:CG	2.17	0.49
1:B:27:ASN:CG	1:B:48:THR:HG22	2.35	0.47
1:B:29:ARG:HH11	1:B:116:LYS:HB3	1.81	0.46
1:A:6:LYS:HE2	1:A:125:ARG:HD2	1.98	0.45
1:B:33:ARG:NH1	2:B:306:HOH:O	2.41	0.44
1:B:197:LYS:HB3	1:B:197:LYS:HE2	1.73	0.43
1:B:29:ARG:NH1	1:B:116:LYS:HB3	2.34	0.43
1:A:27[B]:ASN:HB2	1:A:46:TYR:CZ	2.54	0.42
1:A:15:GLU:HG2	1:A:19:LYS:NZ	2.34	0.42
1:A:182:PRO:HG2	1:A:185:GLN:OE1	2.20	0.41
1:B:243:CYS:HA	1:B:244:PRO:C	2.41	0.41
1:B:6:LYS:HE2	1:B:6:LYS:HB2	1.98	0.40

All (3) symmetry-related close contacts are listed below. The label for Atom-2 includes the symmetry operator and encoded unit-cell translations to be applied.

Atom-1	Atom-2	Interatomic distance (Å)	Clash overlap (Å)
2:A:543:HOH:O	2:B:483:HOH:O[1_655]	1.98	0.22
2:B:471:HOH:O	2:B:499:HOH:O[1_455]	2.10	0.10
2:B:309:HOH:O	2:B:426:HOH:O[2_556]	2.15	0.05

5.3 Torsion angles

5.3.1 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 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	258/269 (96%)	252 (98%)	6 (2%)	0	100	100
1	B	259/269 (96%)	253 (98%)	6 (2%)	0	100	100
All	All	517/538 (96%)	505 (98%)	12 (2%)	0	100	100

There are no Ramachandran outliers to report.

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	221/227 (97%)	221 (100%)	0	100	100
1	B	221/227 (97%)	221 (100%)	0	100	100
All	All	442/454 (97%)	442 (100%)	0	100	100

There are no protein residues with a non-rotameric sidechain to report.

Sometimes sidechains can be flipped to improve hydrogen bonding and reduce clashes. All (1) such sidechains are listed below:

Mol	Chain	Res	Type
1	A	113	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 oligosaccharides 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, 95th 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	257/269 (95%)	0.46	13 (5%) 34 37	5, 11, 23, 42	5 (1%)
1	B	259/269 (96%)	0.85	32 (12%) 9 11	7, 14, 27, 50	2 (0%)
All	All	516/538 (95%)	0.65	45 (8%) 17 19	5, 13, 26, 50	7 (1%)

All (45) RSRZ outliers are listed below:

Mol	Chain	Res	Type	RSRZ
1	B	264	HIS	5.2
1	A	48	THR	4.4
1	B	53	GLY	3.9
1	B	51	THR	3.7
1	A	52	ASP	3.7
1	B	49	ASN	3.5
1	B	48	THR	3.5
1	B	65	THR	3.2
1	B	50	THR	3.1
1	A	53	GLY	3.1
1	B	54	LEU	3.0
1	B	6	LYS	3.0
1	A	36	VAL	2.9
1	B	262	LYS	2.8
1	B	263	GLU	2.8
1	B	29	ARG	2.7
1	B	52	ASP	2.7
1	A	34	LEU	2.6
1	A	69	GLU	2.6
1	B	248	ASN	2.5
1	B	120	ILE	2.5
1	B	12	PRO	2.5
1	A	21	ARG	2.5
1	A	262	LYS	2.5

Continued on next page...

Continued from previous page...

Mol	Chain	Res	Type	RSRZ
1	B	31	VAL	2.4
1	B	37	LYS	2.4
1	B	15	GLU	2.4
1	B	21[A]	ARG	2.4
1	B	23	PRO	2.4
1	B	64	PHE	2.4
1	B	28	ARG	2.4
1	B	26	VAL	2.3
1	B	33	ARG	2.3
1	B	55	PHE	2.3
1	B	34	LEU	2.3
1	B	7	VAL	2.3
1	B	117	VAL	2.3
1	B	27	ASN	2.3
1	A	19	LYS	2.2
1	A	167	ARG	2.2
1	B	69	GLU	2.2
1	A	4	PRO	2.2
1	A	185	GLN	2.1
1	A	35	LYS	2.1
1	B	116	LYS	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.