Full wwPDB X-ray Structure Validation Report (i)

Nov 26, 2024 – 02:13 PM JST

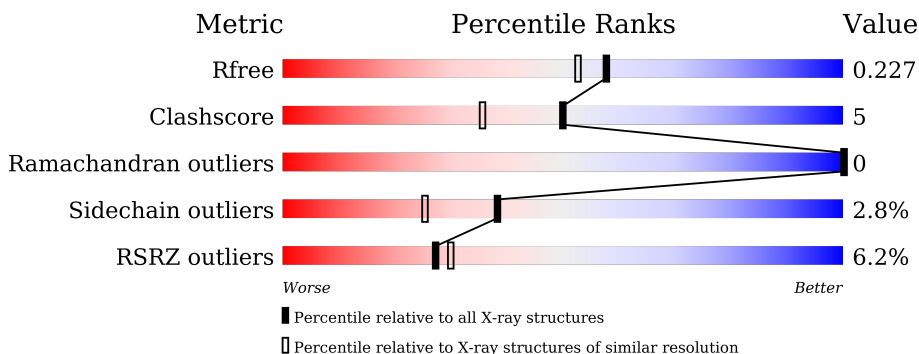|		
PDB ID	:	8JLT
Title	:	Crystal structure of the Green fluorescent protein SET203EF223DA227 variant at pH 7.0
Authors	:	Shin, S.C.
Deposited on	:	2023-06-02
Resolution	:	1.94 Å(reported)

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 1.94 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	Percentile Ranks	Value
Rfree		0.227
Clashscore		5
Ramachandran outliers		0
Sidechain outliers		2.8%
RSRZ outliers		6.2%

Worse   *Better*

▮ Percentile relative to all X-ray structures

▯ Percentile relative to X-ray structures of similar resolution

Metric	Whole archive (#Entries)	Similar resolution (#Entries, resolution range(Å))
R_{free}	164625	1306 (1.94-1.94)
Clashscore	180529	1400 (1.94-1.94)
Ramachandran outliers	177936	1387 (1.94-1.94)
Sidechain outliers	177891	1387 (1.94-1.94)
RSRZ outliers	164620	1306 (1.94-1.94)

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	236	5% 86% 9% 5%
1	B	236	7% 81% 13% • 5%

2 Entry composition ⓘ

There are 2 unique types of molecules in this entry. The entry contains 3865 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 Green fluorescent protein.

Mol	Chain	Residues	Atoms					ZeroOcc	AltConf	Trace
1	A	225	Total	C	N	O	S	0	0	0
			1803	1146	304	347	6			
1	B	225	Total	C	N	O	S	0	0	0
			1803	1146	304	347	6			

There are 26 discrepancies between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	Comment	Reference
A	64	LEU	PHE	engineered mutation	UNP P42212
A	66	CRO	SER	chromophore	UNP P42212
A	66	CRO	TYR	chromophore	UNP P42212
A	66	CRO	GLY	chromophore	UNP P42212
A	80	ARG	GLN	engineered mutation	UNP P42212
A	147	ASP	SER	engineered mutation	UNP P42212
A	149	GLN	ASN	engineered mutation	UNP P42212
A	163	ALA	VAL	engineered mutation	UNP P42212
A	175	GLY	SER	engineered mutation	UNP P42212
A	202	PHE	SER	engineered mutation	UNP P42212
A	204	GLU	GLN	engineered mutation	UNP P42212
A	206	THR	ALA	engineered mutation	UNP P42212
A	223	ASP	PHE	engineered mutation	UNP P42212
B	64	LEU	PHE	engineered mutation	UNP P42212
B	66	CRO	SER	chromophore	UNP P42212
B	66	CRO	TYR	chromophore	UNP P42212
B	66	CRO	GLY	chromophore	UNP P42212
B	80	ARG	GLN	engineered mutation	UNP P42212
B	147	ASP	SER	engineered mutation	UNP P42212
B	149	GLN	ASN	engineered mutation	UNP P42212
B	163	ALA	VAL	engineered mutation	UNP P42212
B	175	GLY	SER	engineered mutation	UNP P42212
B	202	PHE	SER	engineered mutation	UNP P42212
B	204	GLU	GLN	engineered mutation	UNP P42212
B	206	THR	ALA	engineered mutation	UNP P42212

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Chain	Residue	Modelled	Actual	Comment	Reference
B	223	ASP	PHE	engineered mutation	UNP P42212

• Molecule 2 is water.

Mol	Chain	Residues	Atoms		ZeroOcc	AltConf
2	A	121	Total 121	O 121	0	0
2	B	138	Total 138	O 138	0	0

3 Residue-property plots ⓘ

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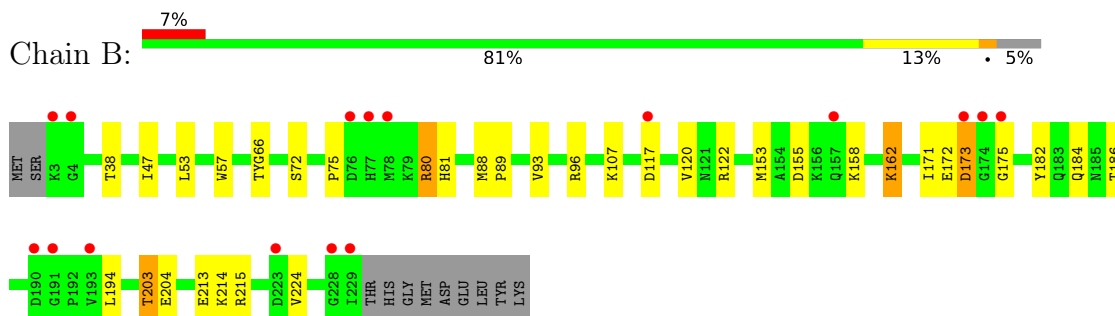 and red = 3 or more. A red dot above a residue indicates a poor fit to the electron density (RSRZ > 2). Stretches of 2 or more consecutive residues without any outlier are shown as a green connector. Residues present in the sample, but not in the model, are shown in grey.

• Molecule 1: Green fluorescent protein

Chain A:

• Molecule 1: Green fluorescent protein

Chain B:
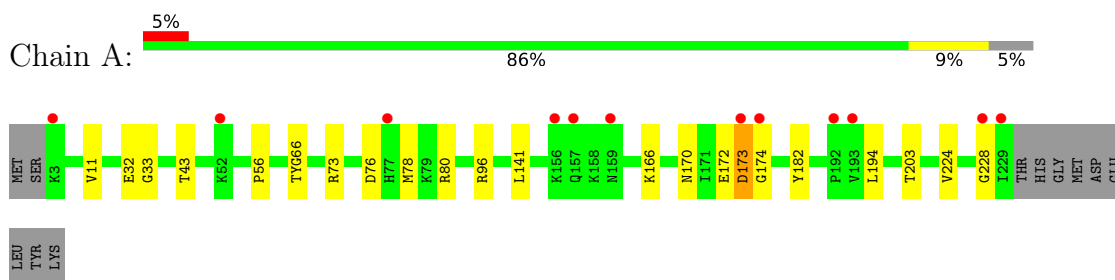
4 Data and refinement statistics ⓘ

Property	Value	Source
Space group	P 1 21 1	Depositor
Cell constants a, b, c, α, β, γ	38.18Å 122.88Å 58.70Å 90.00° 101.06° 90.00°	Depositor
Resolution (Å)	33.38 – 1.94 33.38 – 1.94	Depositor EDS
% Data completeness (in resolution range)	92.4 (33.38-1.94) 92.3 (33.38-1.94)	Depositor EDS
R_{merge}	0.08	Depositor
R_{sym}	(Not available)	Depositor
$< I/\sigma(I) >$ [1]	3.38 (at 1.95Å)	Xtriage
Refinement program	PHENIX (1.19.2_4158: ???)	Depositor
R, R_{free}	0.201 , 0.229 0.208 , 0.227	Depositor DCC
R_{free} test set	37027 reflections (5.54%)	wwPDB-VP
Wilson B-factor (Å^2)	20.6	Xtriage
Anisotropy	0.884	Xtriage
Bulk solvent k_{sol}(e/Å^3), B_{sol}(Å^2)	0.36 , 35.9	EDS
L-test for twinning[2]	$< |L| > = 0.48, < L^2 > = 0.31$	Xtriage
Estimated twinning fraction	No twinning to report.	Xtriage
F_o,F_c correlation	0.94	EDS
Total number of atoms	3865	wwPDB-VP
Average B, all atoms (Å^2)	28.0	wwPDB-VP

Xtriage's analysis on translational NCS is as follows: *The largest off-origin peak in the Patterson function is 7.16% of the height of the origin peak. No significant pseudotranslation is detected.*

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[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 ⓘ

Bond lengths and bond angles in the following residue types are not validated in this section: CRO

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	0.44	0/1821	0.66	0/2461
1	B	0.42	0/1821	0.69	0/2461
All	All	0.43	0/3642	0.67	0/4922

There are no bond length outliers.

There are no bond angle outliers.

There are no chirality outliers.

There are no planarity outliers.

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	1803	0	1733	16	0
1	B	1803	0	1733	25	0
2	A	121	0	0	3	0
2	B	138	0	0	2	0
All	All	3865	0	3466	37	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 5.

All (37) 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:171:ILE:CG1	1:B:175:GLY:O	2.22	0.87
1:B:72:SER:HA	1:B:224:VAL:HG23	1.59	0.84
1:A:174:GLY:HA3	1:B:75:PRO:HB3	1.68	0.76
1:B:171:ILE:CB	1:B:175:GLY:O	2.37	0.72
1:A:32:GLU:HG3	2:A:333:HOH:O	1.89	0.72
1:B:203:THR:HB	1:B:224:VAL:HG12	1.72	0.70
1:B:171:ILE:HB	1:B:175:GLY:O	1.94	0.67
1:B:107:LYS:NZ	2:B:302:HOH:O	2.34	0.61
1:B:213:GLU:OE2	1:B:215:ARG:HD3	2.03	0.59
1:B:80:ARG:O	1:B:194:LEU:HG	2.07	0.54
1:A:203:THR:HG22	1:A:224:VAL:HG12	1.89	0.54
1:B:47:ILE:HD13	1:B:215:ARG:NH2	2.23	0.53
1:B:155:ASP:HB2	1:B:162:LYS:HG2	1.93	0.50
1:A:174:GLY:HA3	1:B:75:PRO:CB	2.39	0.49
1:B:80:ARG:HG2	1:B:81:HIS:CD2	2.48	0.48
1:A:172:GLU:O	1:A:173:ASP:HB2	2.13	0.48
1:A:170:ASN:ND2	1:B:38:THR:O	2.45	0.47
1:B:88:MET:HB3	1:B:89:PRO:HA	1.97	0.47
1:B:172:GLU:O	1:B:173:ASP:CB	2.61	0.47
1:B:153:MET:HE2	1:B:162:LYS:HE3	1.98	0.46
1:B:172:GLU:O	1:B:173:ASP:HB2	2.16	0.46
1:A:32:GLU:CG	2:A:333:HOH:O	2.57	0.45
1:B:155:ASP:OD2	1:B:158:LYS:HD2	2.17	0.45
1:A:73:ARG:HH11	1:A:73:ARG:HD3	1.69	0.43
1:B:96:ARG:HA	1:B:182:TYR:O	2.17	0.43
1:A:172:GLU:O	1:A:173:ASP:CB	2.67	0.42
1:B:107:LYS:HG3	2:B:348:HOH:O	2.18	0.42
1:A:174:GLY:HA2	1:B:75:PRO:HA	2.02	0.42
1:A:33:GLY:HA3	1:A:43:THR:O	2.19	0.42
1:A:56:PRO:HG2	1:A:141:LEU:HD12	2.01	0.42
1:A:78:MET:HE1	1:A:228:GLY:N	2.35	0.41
1:A:11:VAL:HG12	2:A:341:HOH:O	2.20	0.41
1:A:80:ARG:O	1:A:194:LEU:HD23	2.20	0.41
1:B:120:VAL:HG12	1:B:122:ARG:HG3	2.03	0.41
1:B:53:LEU:HD22	1:B:57:TRP:CE2	2.55	0.41
1:A:96:ARG:HA	1:A:182:TYR:O	2.20	0.40
1:B:93:VAL:HB	1:B:186:THR:HG22	2.02	0.40

There are no symmetry-related clashes.

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	220/236 (93%)	215 (98%)	5 (2%)	0	100	100
1	B	220/236 (93%)	216 (98%)	4 (2%)	0	100	100
All	All	440/472 (93%)	431 (98%)	9 (2%)	0	100	100

There are no Ramachandran outliers to report.

5.3.2 Protein sidechains ⓘ

In the following table, the Percentiles column shows the percent sidechain outliers of the chain as a percentile score with respect to all 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	193/205 (94%)	190 (98%)	3 (2%)	58	49
1	B	193/205 (94%)	185 (96%)	8 (4%)	26	12
All	All	386/410 (94%)	375 (97%)	11 (3%)	38	25

All (11) residues with a non-rotameric sidechain are listed below:

Mol	Chain	Res	Type
1	A	76	ASP
1	A	166	LYS
1	A	173	ASP
1	B	80	ARG
1	B	117	ASP
1	B	162	LYS
1	B	173	ASP
1	B	184	GLN

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Mol	Chain	Res	Type
1	B	203	THR
1	B	204	GLU
1	B	214	LYS

Sometimes sidechains can be flipped to improve hydrogen bonding and reduce clashes. All (3) such sidechains are listed below:

Mol	Chain	Res	Type
1	A	184	GLN
1	B	77	HIS
1	B	139	HIS

5.3.3 RNA ⓘ

There are no RNA molecules in this entry.

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

2 non-standard protein/DNA/RNA residues are modelled in this entry.

In the following table, the Counts columns list the number of bonds (or angles) for which Mogul statistics could be retrieved, the number of bonds (or angles) that are observed in the model and the number of bonds (or 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 length (or angle) is the number of standard deviations the observed value is removed from the expected value. A bond length (or angle) with $|Z| > 2$ is considered an outlier worth inspection. RMSZ is the root-mean-square of all Z scores of the bond lengths (or angles).

Mol	Type	Chain	Res	Link	Bond lengths			Bond angles		
					Counts	RMSZ	$\#|Z| > 2$	Counts	RMSZ	$\#|Z| > 2$
1	CRO	B	66	1	23,23,24	6.06	10 (43%)	30,32,34	3.20	10 (33%)
1	CRO	A	66	1	23,23,24	6.06	10 (43%)	30,32,34	3.43	10 (33%)

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	CRO	B	66	1	-	1/12/31/32	0/2/2/2
1	CRO	A	66	1	-	1/12/31/32	0/2/2/2

All (20) bond length outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(Å)	Ideal(Å)
1	A	66	CRO	CB2-CA2	20.79	1.52	1.35
1	B	66	CRO	CB2-CA2	20.79	1.52	1.35
1	A	66	CRO	CA2-C2	-12.81	1.36	1.48
1	B	66	CRO	CA2-C2	-12.76	1.36	1.48
1	A	66	CRO	OG1-CB1	-10.73	1.13	1.43
1	B	66	CRO	OG1-CB1	-10.71	1.13	1.43
1	A	66	CRO	O2-C2	6.31	1.36	1.23
1	B	66	CRO	O2-C2	6.29	1.36	1.23
1	B	66	CRO	CA2-N2	-4.83	1.28	1.38
1	A	66	CRO	CA2-N2	-4.78	1.28	1.38
1	B	66	CRO	C2-N3	-4.60	1.29	1.39
1	A	66	CRO	C2-N3	-4.59	1.29	1.39
1	A	66	CRO	C1-N3	-4.46	1.29	1.37
1	B	66	CRO	C1-N3	-4.46	1.29	1.37
1	B	66	CRO	CG2-CB2	2.56	1.51	1.46
1	A	66	CRO	CG2-CB2	2.56	1.51	1.46
1	B	66	CRO	C1-N2	-2.39	1.28	1.32
1	A	66	CRO	C1-N2	-2.37	1.28	1.32
1	B	66	CRO	CA3-N3	-2.36	1.42	1.47
1	A	66	CRO	CA3-N3	-2.35	1.42	1.47

All (20) bond angle outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(°)	Ideal(°)
1	B	66	CRO	CA2-C2-N3	9.30	107.77	103.37
1	A	66	CRO	CA2-C2-N3	9.29	107.77	103.37
1	B	66	CRO	O2-C2-CA2	-7.65	126.66	130.96
1	A	66	CRO	O2-C2-CA2	-7.54	126.73	130.96
1	A	66	CRO	C1-CA1-N1	7.53	122.18	109.96
1	B	66	CRO	CA2-N2-C1	5.80	110.05	105.77
1	A	66	CRO	CA2-N2-C1	5.76	110.02	105.77
1	A	66	CRO	CG2-CB2-CA2	-5.48	123.22	129.94
1	B	66	CRO	CG2-CB2-CA2	-5.45	123.27	129.94
1	B	66	CRO	C2-CA2-N2	-4.85	105.54	108.93
1	A	66	CRO	C2-CA2-N2	-4.83	105.55	108.93
1	A	66	CRO	N3-C1-N2	-3.98	108.70	111.45
1	B	66	CRO	N3-C1-N2	-3.97	108.71	111.45
1	A	66	CRO	O3-C3-CA3	-3.70	115.23	126.39
1	B	66	CRO	O3-C3-CA3	-3.69	115.25	126.39
1	B	66	CRO	C1-CA1-N1	3.26	115.25	109.96
1	A	66	CRO	OG1-CB1-CA1	3.15	115.79	109.04
1	B	66	CRO	OG1-CB1-CA1	3.14	115.77	109.04

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Mol	Chain	Res	Type	Atoms	Z	Observed(^{o})	Ideal(^{o})
1	A	66	CRO	CA1-C1-N3	2.59	127.86	124.75
1	B	66	CRO	CA1-C1-N3	2.57	127.82	124.75

There are no chirality outliers.

All (2) torsion outliers are listed below:

Mol	Chain	Res	Type	Atoms
1	A	66	CRO	C1-CA1-CB1-CG1
1	B	66	CRO	C1-CA1-CB1-CG1

There are no ring outliers.

No monomer is involved in short contacts.

5.5 Carbohydrates ⓘ

There are no oligosaccharides in this entry.

5.6 Ligand geometry ⓘ

There are no ligands in this entry.

5.7 Other polymers ⓘ

There are no such residues in this entry.

5.8 Polymer linkage issues ⓘ

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	224/236 (94%)	0.26	12 (5%)	32	36	16, 26, 50, 63	0
1	B	224/236 (94%)	0.27	16 (7%)	23	26	15, 25, 50, 63	0
All	All	448/472 (94%)	0.27	28 (6%)	27	30	15, 26, 50, 63	0

All (28) RSRZ outliers are listed below:

Mol	Chain	Res	Type	RSRZ
1	B	175	GLY	6.0
1	A	174	GLY	5.0
1	B	174	GLY	4.2
1	A	229	ILE	4.1
1	B	3	LYS	4.0
1	B	228	GLY	4.0
1	B	229	ILE	3.9
1	B	191	GLY	3.9
1	B	173	ASP	3.7
1	A	77	HIS	3.6
1	B	77	HIS	3.5
1	B	117	ASP	3.2
1	B	223	ASP	2.7
1	A	193	VAL	2.6
1	A	3	LYS	2.6
1	B	190	ASP	2.6
1	A	52	LYS	2.5
1	A	159	ASN	2.5
1	A	192	PRO	2.4
1	B	193	VAL	2.3
1	B	157	GLN	2.3
1	A	173	ASP	2.3
1	A	157	GLN	2.2
1	B	4	GLY	2.2

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Mol	Chain	Res	Type	RSRZ
1	B	78	MET	2.2
1	A	228	GLY	2.1
1	B	76	ASP	2.0
1	A	156	LYS	2.0

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

In the following table, the Atoms column lists the number of modelled atoms in the group and the number defined in the chemical component dictionary. The B-factors column lists the minimum, median, 95th percentile and maximum values of B factors of atoms in the group. The column labelled 'Q< 0.9' lists the number of atoms with occupancy less than 0.9.

Mol	Type	Chain	Res	Atoms	RSCC	RSR	B-factors(Å^2)	Q<0.9
1	CRO	A	66	22/23	0.88	0.11	17,22,24,25	0
1	CRO	B	66	22/23	0.90	0.10	17,22,24,25	0

6.3 Carbohydrates ⓘ

There are no monosaccharides in this entry.

6.4 Ligands ⓘ

There are no ligands in this entry.

6.5 Other polymers ⓘ

There are no such residues in this entry.