



# Full wwPDB X-ray Structure Validation Report

Feb 28, 2014 – 05:32 AM GMT

PDB ID : 2B3P  
Title : Crystal structure of a superfolder green fluorescent protein  
Authors : Pedelacq, J.D.; Cabantous, S.; Tran, T.H.; Terwilliger, T.C.; Waldo, G.S.  
Deposited on : 2005-09-20  
Resolution : 1.40 Å(reported)

This is a full wwPDB validation report for a publicly released PDB entry.  
We welcome your comments at [validation@mail.wwpdb.org](mailto:validation@mail.wwpdb.org)  
A user guide is available at <http://wwpdb.org/ValidationPDFNotes.html>

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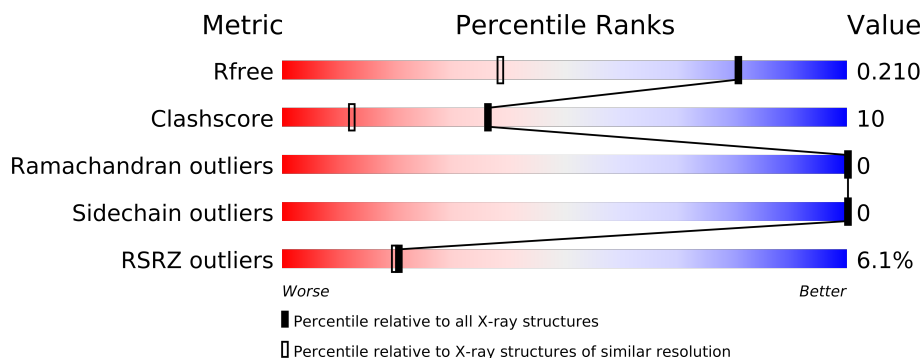
The following versions of software and data (see [references](#)) were used in the production of this report:

MolProbity : 4.02b-467  
Mogul : 1.15 2013  
Xtriage (Phenix) : dev-1323  
EDS : stable22639  
Percentile statistics : 21963  
Refmac : 5.8.0049  
CCP4 : 6.3.0 (Settle)  
Ideal geometry (proteins) : Engh & Huber (2001)  
Ideal geometry (DNA, RNA) : Parkinson et. al. (1996)  
Validation Pipeline (wwPDB-VP) : stable22683

# 1 Overall quality at a glance

The reported resolution of this entry is 1.40 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))
$R_{free}$	66092	1097 (1.42-1.38)
Clashscore	79885	1246 (1.42-1.38)
Ramachandran outliers	78287	1206 (1.42-1.38)
Sidechain outliers	78261	1205 (1.42-1.38)
RSRZ outliers	66119	1097 (1.42-1.38)

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 on the lower bar indicate the fraction of residues that contain outliers for  $\geq 3$ , 2, 1 and 0 types of geometric quality criteria. The upper red bar (where present) indicates the fraction of residues that have poor fit to the electron density.

Mol	Chain	Length	Quality of chain
1	A	244	

The following table lists non-polymeric compounds that are outliers for geometric or electron-density-fit criteria:

Mol	Type	Chain	Res	Geometry	Electron density
2	CD	A	806	-	X
2	CD	A	807	-	X
2	CD	A	808	-	X
2	CD	A	809	-	X
3	ACY	A	701	-	X
3	ACY	A	704	-	X
3	ACY	A	705	-	X
3	ACY	A	707	-	X

## 2 Entry composition

There are 4 unique types of molecules in this entry. The entry contains 2165 atoms, of which 0 are hydrogen and 0 are deuterium.

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	229	Total	C	N	O	S	0	0	0
			1804	1140	310	349	5			

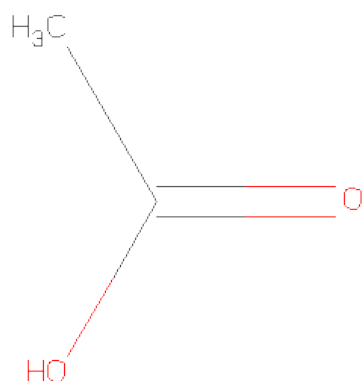
There are 22 discrepancies between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	Comment	Reference
A	30	ARG	SER	ENGINEERED	UNP P42212
A	39	ASN	TYR	ENGINEERED	UNP P42212
A	64	LEU	PHE	ENGINEERED	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	UNP P42212
A	99	SER	PHE	ENGINEERED	UNP P42212
A	105	THR	ASN	ENGINEERED	UNP P42212
A	145	PHE	TYR	ENGINEERED	UNP P42212
A	153	THR	MET	ENGINEERED	UNP P42212
A	163	ALA	VAL	ENGINEERED	UNP P42212
A	171	VAL	ILE	ENGINEERED	UNP P42212
A	206	VAL	ALA	ENGINEERED	UNP P42212
A	239	GLY	-	EXPRESSION TAG	UNP P42212
A	240	SER	-	EXPRESSION TAG	UNP P42212
A	241	HIS	-	EXPRESSION TAG	UNP P42212
A	242	HIS	-	EXPRESSION TAG	UNP P42212
A	243	HIS	-	EXPRESSION TAG	UNP P42212
A	244	HIS	-	EXPRESSION TAG	UNP P42212
A	245	HIS	-	EXPRESSION TAG	UNP P42212
A	246	HIS	-	EXPRESSION TAG	UNP P42212

- Molecule 2 is CADMIUM ION (three-letter code: CD) (formula: Cd).

Mol	Chain	Residues	Atoms		ZeroOcc	AltConf
2	A	9	Total	Cd	0	0
			9	9		

- Molecule 3 is ACETIC ACID (three-letter code: ACY) (formula: C<sub>2</sub>H<sub>4</sub>O<sub>2</sub>).



Mol	Chain	Residues	Atoms			ZeroOcc	AltConf
3	A	1	Total	C	O	0	0
			4	2	2		
3	A	1	Total	C	O	0	0
			4	2	2		
3	A	1	Total	C	O	0	0
			4	2	2		
3	A	1	Total	C	O	0	0
			4	2	2		
3	A	1	Total	C	O	0	0
			4	2	2		

- Molecule 4 is water.

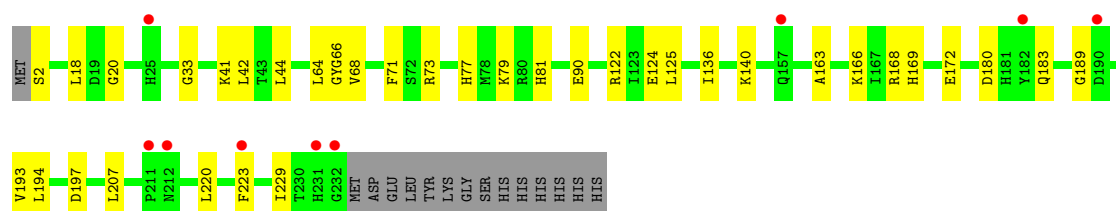
Mol	Chain	Residues	Atoms		ZeroOcc	AltConf
4	A	328	Total	O	0	0
			328	328		

### 3 Residue-property plots

These plots are drawn for all protein, RNA and DNA chains in the entry. The first graphic for a chain summarises the proportions of errors 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: 



## 4 Data and refinement statistics

Property	Value	Source
Space group	P 31 2 1	Depositor
Cell constants a, b, c, $\alpha$ , $\beta$ , $\gamma$	88.46Å 88.46Å 69.35Å 90.00° 90.00° 120.00°	Depositor
Resolution (Å)	7.99 – 1.40 21.25 – 1.33	Depositor EDS
% Data completeness (in resolution range)	93.2 (7.99-1.40) 83.5 (21.25-1.33)	Depositor EDS
$R_{merge}$	(Not available)	Depositor
$R_{sym}$	(Not available)	Depositor
$\langle I/\sigma(I) \rangle$ <sup>1</sup>	0.99 (at 1.33Å)	Xtriage
Refinement program	CNS 1.1	Depositor
R, $R_{free}$	0.206 , 0.218 0.211 , 0.210	Depositor DCC
$R_{free}$ test set	4059 reflections (7.07%)	DCC
Wilson B-factor (Å <sup>2</sup> )	11.5	Xtriage
Anisotropy	0.604	Xtriage
Bulk solvent $k_{sol}$ (e/Å <sup>3</sup> ), $B_{sol}$ (Å <sup>2</sup> )	0.48 , 45.0	EDS
Estimated twinning fraction	0.026 for -h,-k,l	Xtriage
L-test for twinning	$\langle  L  \rangle = 0.49$ , $\langle L^2 \rangle = 0.32$	Xtriage
Outliers	1 of 60830 reflections (0.002%)	Xtriage
$F_o, F_c$ correlation	0.94	EDS
Total number of atoms	2165	wwPDB-VP
Average B, all atoms (Å <sup>2</sup> )	16.0	wwPDB-VP

Xtriage's analysis on translational NCS is as follows: *The largest off-origin peak in the Patterson function is 6.18% of the height of the origin peak. No significant pseudotranslation is detected.*

<sup>1</sup>Intensities estimated from amplitudes.

## 5 Model quality

### 5.1 Standard geometry

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

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	0.33	0/1821	0.67	1/2466 (0.0%)

There are no bond length outliers.

All (1) bond angle outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed( $^{\circ}$ )	Ideal( $^{\circ}$ )
1	A	122	ARG	NE-CZ-NH2	6.10	123.35	120.30

There are no chirality outliers.

There are no planarity outliers.

### 5.2 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 hydrogens added by MolProbity. The Clashes column lists the number of clashes within the asymmetric unit, and the number in parentheses is this value normalized per 1000 atoms of the molecule in the chain. The Symm-Clashes column gives symmetry related clashes, in the same way as for the Clashes column.

Mol	Chain	Non-H	H(model)	H(added)	Clashes	Symm-Clashes
1	A	1804	0	1728	35	0
2	A	9	0	0	0	0
3	A	24	0	18	1	0
4	A	328	0	0	3	0
All	All	2165	0	1746	35	0

Clashscore is defined as the number of clashes calculated for the entry per 1000 atoms (including hydrogens) of the entry. The overall clashscore for this entry is 10.

All (35) close contacts within the same asymmetric unit are listed below.

Atom-1	Atom-2	Distance(Å)	Clash(Å)
1:A:64:LEU:C	1:A:66:CRO:H2	1.45	1.06
1:A:81:HIS:HD2	1:A:197:ASP:H	1.26	0.83
1:A:193:VAL:HG12	1:A:194:LEU:HD12	1.64	0.80
1:A:66:CRO:C3	1:A:68:VAL:CA	2.61	0.79
1:A:77:HIS:HE1	1:A:229:ILE:O	1.70	0.72
1:A:66:CRO:CA3	1:A:68:VAL:N	2.54	0.70
1:A:81:HIS:CD2	1:A:197:ASP:H	2.12	0.68
1:A:33:GLY:HA3	1:A:44:LEU:HD23	1.76	0.67
1:A:64:LEU:C	1:A:66:CRO:CA1	2.62	0.65
1:A:42:LEU:HD21	1:A:71:PHE:CG	2.38	0.58
1:A:64:LEU:CA	1:A:66:CRO:H2	2.19	0.56
1:A:18:LEU:HD23	1:A:18:LEU:C	2.29	0.53
1:A:33:GLY:CA	1:A:44:LEU:HD23	2.39	0.50
1:A:207:LEU:CD2	1:A:220:LEU:HD13	2.41	0.50
1:A:193:VAL:HG12	1:A:194:LEU:CD1	2.39	0.50
1:A:140:LYS:O	1:A:172:GLU:HG2	2.12	0.49
1:A:77:HIS:CE1	1:A:229:ILE:O	2.59	0.49
1:A:2:SER:HB3	1:A:79:LYS:HB3	1.94	0.48
1:A:124:GLU:HG2	4:A:1046:HOH:O	2.15	0.47
1:A:125:LEU:C	1:A:125:LEU:HD23	2.35	0.47
1:A:136:ILE:HD12	1:A:136:ILE:N	2.31	0.46
1:A:207:LEU:HD23	1:A:220:LEU:HD13	1.98	0.45
1:A:41:LYS:HB2	1:A:223:PHE:CE2	2.52	0.44
1:A:166:LYS:HG2	1:A:180:ASP:OD1	2.17	0.44
1:A:169:HIS:HD2	4:A:816:HOH:O	2.01	0.43
1:A:64:LEU:CA	1:A:66:CRO:N1	2.72	0.43
1:A:168:ARG:HH21	3:A:706:ACY:H3	1.83	0.42
1:A:41:LYS:HB2	1:A:223:PHE:HE2	1.85	0.42
1:A:193:VAL:C	1:A:194:LEU:HD12	2.39	0.41
1:A:163:ALA:HB3	1:A:183:GLN:HB3	2.02	0.41
1:A:90:GLU:CD	1:A:189:GLY:HA3	2.40	0.41
1:A:42:LEU:HD21	1:A:71:PHE:CD2	2.56	0.41
1:A:42:LEU:HD21	1:A:71:PHE:CB	2.51	0.41
1:A:20:GLY:HA2	1:A:125:LEU:O	2.21	0.41
1:A:73:ARG:NH2	4:A:1034:HOH:O	2.52	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	224/244 (92%)	222 (99%)	2 (1%)	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	194/213 (91%)	194 (100%)	0	100	100

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

Some sidechains can be flipped to improve hydrogen bonding and reduce clashes. All (7) such sidechains are listed below:

Mol	Chain	Res	Type
1	A	77	HIS
1	A	81	HIS
1	A	135	ASN
1	A	149	ASN
1	A	159	ASN
1	A	169	HIS
1	A	177	GLN

### 5.3.3 RNA ⓘ

There are no RNA chains in this entry.

## 5.4 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 (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	A	66	1	23,23,24	4.38	10 (43%)	30,32,34	2.37	5 (16%)

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

All (10) bond length outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(Å)	Ideal(Å)
1	A	66	CRO	O3-C3	15.15	1.21	1.11
1	A	66	CRO	CB2-CA2	6.26	1.39	1.35
1	A	66	CRO	CD1-CG2	5.70	1.50	1.39
1	A	66	CRO	CE1-CZ	5.60	1.50	1.38
1	A	66	CRO	CE2-CZ	5.44	1.50	1.38
1	A	66	CRO	CD2-CG2	4.47	1.48	1.39
1	A	66	CRO	OH-CZ	-4.41	1.25	1.37
1	A	66	CRO	C1-N3	3.88	1.44	1.37
1	A	66	CRO	C2-N3	2.32	1.44	1.39
1	A	66	CRO	CG2-CB2	-2.10	1.42	1.46

All (5) bond angle outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(°)	Ideal(°)
1	A	66	CRO	CA3-N3-C2	10.33	129.88	123.46
1	A	66	CRO	N3-C1-N2	3.96	114.41	111.51
1	A	66	CRO	CA1-C1-N3	-2.69	121.79	124.54
1	A	66	CRO	CB1-CA1-N1	-2.33	106.18	109.22

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Mol	Chain	Res	Type	Atoms	Z	Observed(°)	Ideal(°)
1	A	66	CRO	C1-CA1-N1	-2.13	104.62	109.07

There are no chirality outliers.

There are no torsion outliers.

There are no ring outliers.

## 5.5 Carbohydrates

There are no carbohydrates in this entry.

## 5.6 Ligand geometry

Of 15 ligands modelled in this entry, 9 are monoatomic - leaving 6 for Mogul analysis.

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	$\# Z  > 2$
3	ACY	A	701	2	3,3,3	0.76	0	3,3,3	2.47	2 (66%)
3	ACY	A	703	2	3,3,3	0.90	0	3,3,3	2.38	2 (66%)
3	ACY	A	704	2	3,3,3	0.74	0	3,3,3	2.47	2 (66%)
3	ACY	A	705	2	3,3,3	0.73	0	3,3,3	2.46	2 (66%)
3	ACY	A	706	2	3,3,3	0.74	0	3,3,3	2.45	2 (66%)
3	ACY	A	707	2	3,3,3	0.76	0	3,3,3	2.46	2 (66%)

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
3	ACY	A	701	2	-	0/0/0/0	0/0/0/0
3	ACY	A	703	2	-	0/0/0/0	0/0/0/0

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Mol	Type	Chain	Res	Link	Chirals	Torsions	Rings
3	ACY	A	704	2	-	0/0/0/0	0/0/0/0
3	ACY	A	705	2	-	0/0/0/0	0/0/0/0
3	ACY	A	706	2	-	0/0/0/0	0/0/0/0
3	ACY	A	707	2	-	0/0/0/0	0/0/0/0

There are no bond length outliers.

All (12) bond angle outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(°)	Ideal(°)
3	A	701	ACY	OXT-C-CH3	3.11	129.79	115.43
3	A	705	ACY	OXT-C-CH3	3.11	129.77	115.43
3	A	707	ACY	OXT-C-CH3	3.10	129.75	115.43
3	A	704	ACY	OXT-C-CH3	3.10	129.75	115.43
3	A	706	ACY	OXT-C-CH3	3.06	129.57	115.43
3	A	703	ACY	OXT-C-CH3	3.01	129.34	115.43
3	A	704	ACY	O-C-CH3	-2.94	109.25	122.06
3	A	706	ACY	O-C-CH3	-2.93	109.30	122.06
3	A	701	ACY	O-C-CH3	-2.92	109.33	122.06
3	A	705	ACY	O-C-CH3	-2.91	109.38	122.06
3	A	707	ACY	O-C-CH3	-2.90	109.42	122.06
3	A	703	ACY	O-C-CH3	-2.79	109.89	122.06

There are no chirality outliers.

There are no torsion outliers.

There are no ring outliers.

## 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, 95<sup>th</sup> 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 <sup>2</sup> )	Q<0.9
1	A	229/244 (93%)	0.33	9 (3%) 37 39	7, 13, 23, 28	0

All (9) RSRZ outliers are listed below:

Mol	Chain	Res	Type	RSRZ
1	A	212	ASN	4.9
1	A	211	PRO	4.0
1	A	182	TYR	3.4
1	A	223	PHE	3.4
1	A	190	ASP	3.1
1	A	232	GLY	2.7
1	A	25	HIS	2.5
1	A	231	HIS	2.5
1	A	157	GLN	2.2

### 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. LLDF column lists the quality of electron density of the group with respect to its neighbouring residues in protein, DNA or RNA chains. The B-factors column lists the minimum, median, 95<sup>th</sup> 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	RSR	LLDF	B-factors(Å <sup>2</sup> )	Q<0.9
1	CRO	A	66	22/23	0.12	0.89	6,9,14,17	0

### 6.3 Carbohydrates ⓘ

There are no carbohydrates in this entry.

## 6.4 Ligands ⓘ

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. LLDF column lists the quality of electron density of the group with respect to its neighbouring residues in protein, DNA or RNA chains. The B-factors column lists the minimum, median, 95<sup>th</sup> 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	RSR	LLDF	B-factors(Å <sup>2</sup> )	Q<0.9
3	ACY	A	707	4/4	0.37	39.76	38,39,39,39	0
2	CD	A	807	1/1	0.25	13.79	45,45,45,45	0
2	CD	A	809	1/1	0.21	4.40	84,84,84,84	0
3	ACY	A	705	4/4	0.36	4.36	24,24,25,27	0
3	ACY	A	701	4/4	0.35	4.30	41,41,42,42	0
2	CD	A	808	1/1	0.22	3.61	66,66,66,66	0
3	ACY	A	704	4/4	0.19	2.87	28,30,32,32	0
2	CD	A	806	1/1	0.26	2.22	38,38,38,38	0
3	ACY	A	706	4/4	0.15	1.76	28,30,31,31	0
3	ACY	A	703	4/4	0.21	1.62	14,14,16,17	0
2	CD	A	805	1/1	0.12	0.11	34,34,34,34	0
2	CD	A	804	1/1	0.15	-0.03	27,27,27,27	0
2	CD	A	802	1/1	0.04	-1.46	13,13,13,13	0
2	CD	A	803	1/1	0.05	-1.92	14,14,14,14	0
2	CD	A	801	1/1	0.04	-2.01	12,12,12,12	0

## 6.5 Other polymers ⓘ

There are no such residues in this entry.