Full wwPDB X-ray Structure Validation Report (i)

Nov 5, 2023 – 09:27 AM EST

PDB ID	:	5D1C
Title	:	Crystal structure of D233G-Y306F HDAC8 in complex with a tetrapeptide substrate
Authors	:	Decroos, C.; Christianson, N.H.; Gullett, L.E.; Bowman, C.M.; Christianson, K.E.; Deardorff, M.A.; Christianson, D.W.
Deposited on	:	2015-08-04
Resolution	:	1.42 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 (i) 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 (i)) were used in the production of this report:

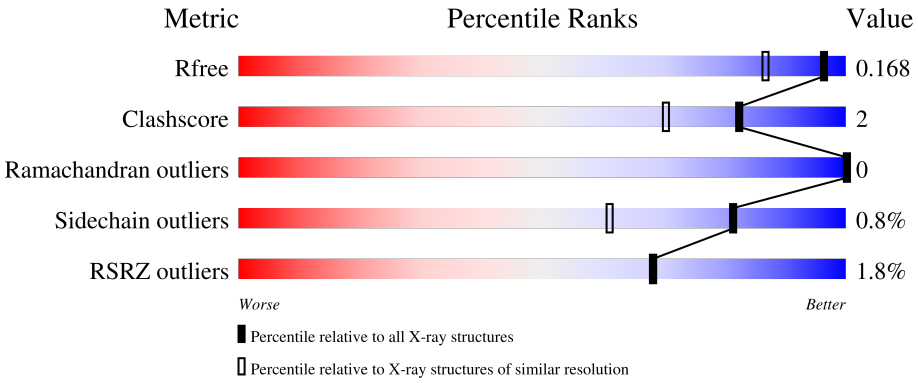
MolProbity	:	4.02b-467
Mogul	:	1.8.5 (274361), CSD as541be (2020)
Xtriage (Phenix)	:	1.13
EDS	:	2.36
Percentile statistics	:	20191225.v01 (using entries in the PDB archive December 25th 2019)
Refmac	:	5.8.0158
CCP4	:	7.0.044 (Gargrove)
Ideal geometry (proteins)	:	Engh & Huber (2001)
Ideal geometry (DNA, RNA)	:	Parkinson et al. (1996)
Validation Pipeline (wwPDB-VP)	:	2.36

1 Overall quality at a glance (i)

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

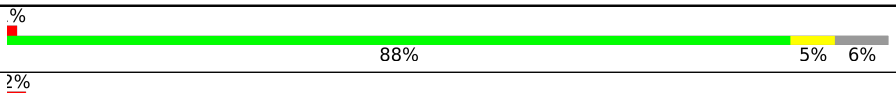
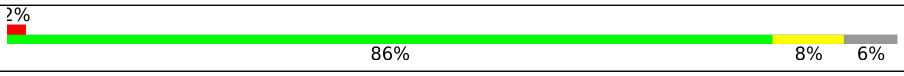
The reported resolution of this entry is 1.42 Å.

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	2579 (1.44-1.40)
Clashscore	141614	2696 (1.44-1.40)
Ramachandran outliers	138981	2632 (1.44-1.40)
Sidechain outliers	138945	2631 (1.44-1.40)
RSRZ outliers	127900	2528 (1.44-1.40)

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	389	
1	B	389	
2	C	6	
2	D	6	

2 Entry composition (i)

There are 6 unique types of molecules in this entry. The entry contains 6892 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 Histone deacetylase 8.

Mol	Chain	Residues	Atoms					ZeroOcc	AltConf	Trace
1	A	364	Total	C	N	O	S	0	20	0
			2922	1874	480	546	22			
1	B	364	Total	C	N	O	S	0	29	0
			3017	1934	495	567	21			

There are 28 discrepancies between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	Comment	Reference
A	233	GLY	ASP	engineered mutation	UNP Q9BY41
A	306	PHE	TYR	engineered mutation	UNP Q9BY41
A	378	ILE	-	expression tag	UNP Q9BY41
A	379	GLU	-	expression tag	UNP Q9BY41
A	380	GLY	-	expression tag	UNP Q9BY41
A	381	ARG	-	expression tag	UNP Q9BY41
A	382	GLY	-	expression tag	UNP Q9BY41
A	383	SER	-	expression tag	UNP Q9BY41
A	384	HIS	-	expression tag	UNP Q9BY41
A	385	HIS	-	expression tag	UNP Q9BY41
A	386	HIS	-	expression tag	UNP Q9BY41
A	387	HIS	-	expression tag	UNP Q9BY41
A	388	HIS	-	expression tag	UNP Q9BY41
A	389	HIS	-	expression tag	UNP Q9BY41
B	233	GLY	ASP	engineered mutation	UNP Q9BY41
B	306	PHE	TYR	engineered mutation	UNP Q9BY41
B	378	ILE	-	expression tag	UNP Q9BY41
B	379	GLU	-	expression tag	UNP Q9BY41
B	380	GLY	-	expression tag	UNP Q9BY41
B	381	ARG	-	expression tag	UNP Q9BY41
B	382	GLY	-	expression tag	UNP Q9BY41
B	383	SER	-	expression tag	UNP Q9BY41
B	384	HIS	-	expression tag	UNP Q9BY41
B	385	HIS	-	expression tag	UNP Q9BY41
B	386	HIS	-	expression tag	UNP Q9BY41

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Chain	Residue	Modelled	Actual	Comment	Reference
B	387	HIS	-	expression tag	UNP Q9BY41
B	388	HIS	-	expression tag	UNP Q9BY41
B	389	HIS	-	expression tag	UNP Q9BY41

- Molecule 2 is a protein called HDAC8 Fluor de Lys tetrapeptide substrate.

Mol	Chain	Residues	Atoms				ZeroOcc	AltConf	Trace
2	C	6	Total	C	N	O	0	0	0
			61	40	12	9			
2	D	6	Total	C	N	O	0	0	0
			61	40	12	9			

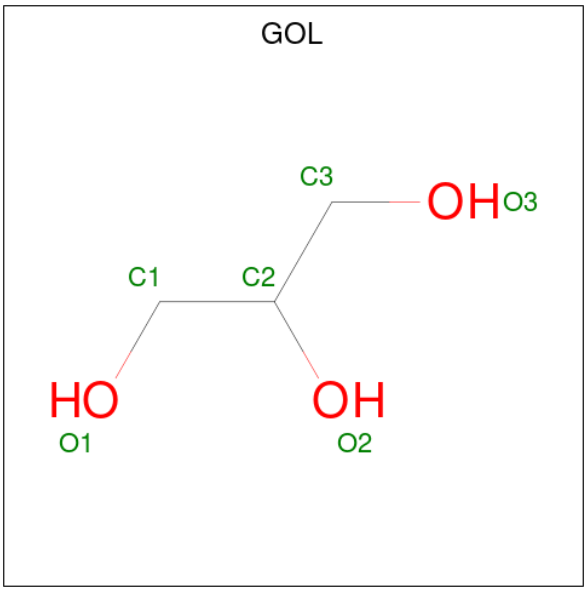
- Molecule 3 is POTASSIUM ION (three-letter code: K) (formula: K).

Mol	Chain	Residues	Atoms		ZeroOcc	AltConf
3	A	2	Total	K	0	0
			2	2		
3	B	2	Total	K	0	0
			2	2		

- Molecule 4 is ZINC ION (three-letter code: ZN) (formula: Zn).

Mol	Chain	Residues	Atoms		ZeroOcc	AltConf
4	A	1	Total	Zn	0	0
			1	1		
4	B	1	Total	Zn	0	0
			1	1		

- Molecule 5 is GLYCEROL (three-letter code: GOL) (formula: $C_3H_8O_3$).



Mol	Chain	Residues	Atoms	ZeroOcc	AltConf
5	B	1	Total C O 6 3 3	0	0

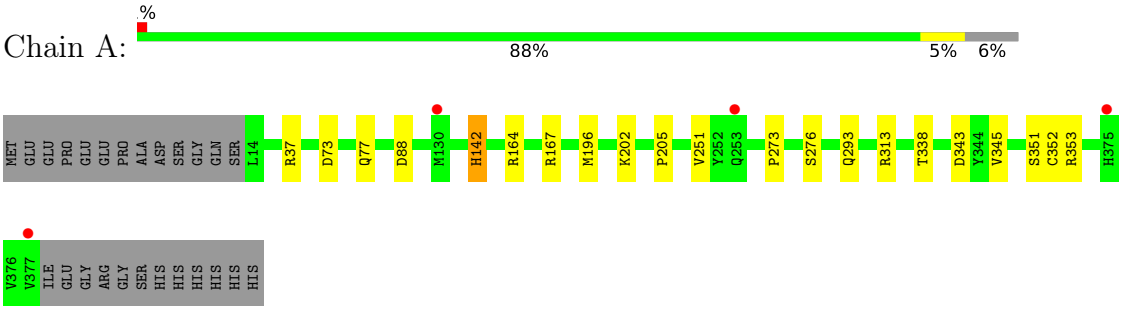
- Molecule 6 is water.

Mol	Chain	Residues	Atoms	ZeroOcc	AltConf
6	A	399	Total O 401 401	0	3
6	B	390	Total O 394 394	0	6
6	C	7	Total O 7 7	0	0
6	D	15	Total O 17 17	0	2

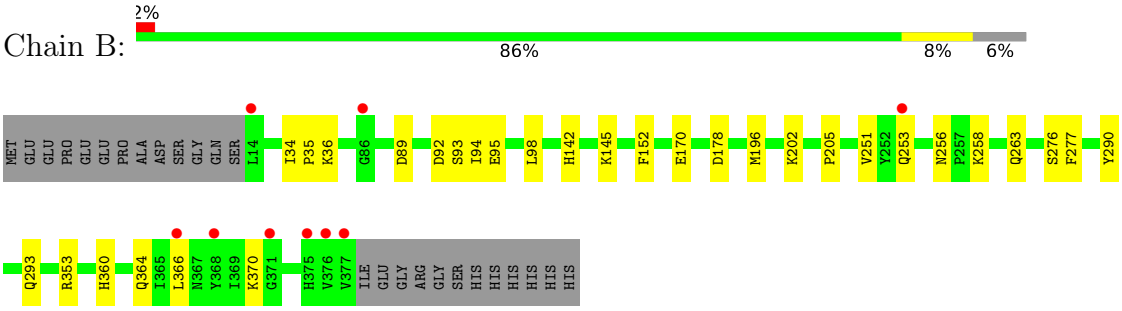
3 Residue-property plots (i)

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: Histone deacetylase 8



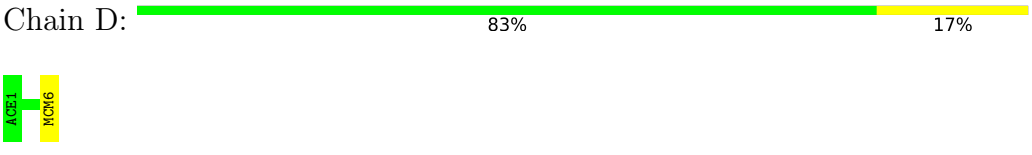
• Molecule 1: Histone deacetylase 8



• Molecule 2: HDAC8 Fluor de Lys tetrapeptide substrate

There are no outlier residues recorded for this chain.

• Molecule 2: HDAC8 Fluor de Lys tetrapeptide substrate



4 Data and refinement statistics (i)

Property	Value	Source
Space group	P 21 21 21	Depositor
Cell constants a, b, c, α, β, γ	83.03Å 97.94Å 104.66Å 90.00° 90.00° 90.00°	Depositor
Resolution (Å)	44.35 – 1.42 44.35 – 1.42	Depositor EDS
% Data completeness (in resolution range)	99.8 (44.35-1.42) 99.8 (44.35-1.42)	Depositor EDS
R_{merge}	0.07	Depositor
R_{sym}	(Not available)	Depositor
$< I/\sigma(I) >$ [1]	1.94 (at 1.42Å)	Xtriage
Refinement program	PHENIX dev_1833	Depositor
R, R_{free}	0.146 , 0.167 0.147 , 0.168	Depositor DCC
R_{free} test set	8011 reflections (5.01%)	wwPDB-VP
Wilson B-factor (Å^2)	15.3	Xtriage
Anisotropy	0.286	Xtriage
Bulk solvent k_{sol}(e/Å^3), B_{sol}(Å^2)	0.36 , 52.2	EDS
L-test for twinning[2]	$< \|L\| > = 0.49$, $< L^2 > = 0.33$	Xtriage
Estimated twinning fraction	No twinning to report.	Xtriage
F_o,F_c correlation	0.98	EDS
Total number of atoms	6892	wwPDB-VP
Average B, all atoms (Å^2)	21.0	wwPDB-VP

Xtriage's analysis on translational NCS is as follows: *The largest off-origin peak in the Patterson function is 4.66% 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

Bond lengths and bond angles in the following residue types are not validated in this section: ZN, ACE, K, ALY, MCM, GOL

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	0/3039	0.63	2/4127 (0.0%)
1	B	1.33	2/3122 (0.1%)	0.95	4/4234 (0.1%)
2	C	0.27	0/22	0.58	0/28
2	D	0.37	0/22	0.63	0/28
All	All	1.00	2/6205 (0.0%)	0.81	6/8417 (0.1%)

All (2) bond length outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(Å)	Ideal(Å)
1	B	89[A]	ASP	CB-CG	49.60	2.56	1.51
1	B	89[B]	ASP	CB-CG	49.60	2.56	1.51

All (6) bond angle outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(^{o})	Ideal(^{o})
1	B	89[A]	ASP	CB-CG-OD2	-29.14	92.07	118.30
1	B	89[B]	ASP	CB-CG-OD2	-29.14	92.07	118.30
1	B	89[A]	ASP	CB-CG-OD1	-14.48	105.27	118.30
1	B	89[B]	ASP	CB-CG-OD1	-14.48	105.27	118.30
1	A	313	ARG	NE-CZ-NH1	-7.68	116.46	120.30
1	A	37	ARG	NE-CZ-NH2	-5.35	117.62	120.30

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	2922	0	2845	14	0
1	B	3017	0	2930	22	0
2	C	61	0	56	0	0
2	D	61	0	56	1	0
3	A	2	0	0	0	0
3	B	2	0	0	0	0
4	A	1	0	0	0	0
4	B	1	0	0	0	0
5	B	6	0	8	0	0
6	A	401	0	0	0	0
6	B	394	0	0	5	0
6	C	7	0	0	0	0
6	D	17	0	0	0	0
All	All	6892	0	5895	30	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 2.

All (30) 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:338[B]:THR:HG23	1:B:94:ILE:HD13	1.71	0.72
1:A:202:LYS:HD2	1:A:276[B]:SER:OG	1.93	0.68
1:B:256[B]:ASN:ND2	6:B:502:HOH:O	2.27	0.68
1:A:73[A]:ASP:OD1	1:A:77:GLN:NE2	2.27	0.67
1:B:92[A]:ASP:HA	6:B:527:HOH:O	1.99	0.63
1:B:93[B]:SER:HB3	1:B:98:LEU:HB2	1.83	0.59
1:A:352[A]:CYS:O	1:B:205:PRO:HG2	2.03	0.58
1:B:202[B]:LYS:HD2	1:B:276[B]:SER:OG	2.04	0.56
1:A:88:ASP:O	1:B:36:LYS:NZ	2.37	0.55
1:B:290:TYR:O	1:B:293:GLN:HG3	2.08	0.53
1:B:145:LYS:HE3	6:B:524:HOH:O	2.07	0.52
1:A:338[B]:THR:HG21	1:B:94:ILE:HG21	1.94	0.50
1:A:351[B]:SER:OG	1:A:353:ARG:HG2	2.11	0.50
1:B:360:HIS:O	1:B:364:GLN:HG2	2.12	0.49
1:B:253:GLN:HG3	6:B:667:HOH:O	2.15	0.47
1:B:277:PHE:O	1:B:353:ARG:NH2	2.49	0.46
1:B:170[B]:GLU:N	6:B:505:HOH:O	2.37	0.46
1:A:205:PRO:O	1:B:353:ARG:HD3	2.17	0.44

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Atom-1	Atom-2	Interatomic distance (Å)	Clash overlap (Å)
1:B:92[A]:ASP:O	1:B:95:GLU:HB3	2.18	0.44
1:A:142:HIS:H	1:A:142:HIS:CD2	2.36	0.43
1:A:196[A]:MET:SD	1:A:251:VAL:HG13	2.59	0.43
1:B:196[A]:MET:SD	1:B:251:VAL:HG13	2.58	0.43
1:A:164:ARG:HD3	1:A:167[B]:ARG:HD2	2.00	0.42
1:A:343[A]:ASP:HB2	1:A:345:VAL:HG22	2.02	0.42
1:B:178:ASP:HB2	1:B:263:GLN:OE1	2.20	0.42
1:B:366:LEU:HG	1:B:370[B]:LYS:HE2	2.01	0.41
1:A:338[B]:THR:CG2	1:B:94:ILE:HG21	2.50	0.41
1:B:34:ILE:N	1:B:35:PRO:HD3	2.35	0.41
1:B:366:LEU:HG	1:B:370[A]:LYS:HE2	2.03	0.40
1:A:273:PRO:HB2	2:D:6:MCM:H9	2.04	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	383/389 (98%)	376 (98%)	7 (2%)	0	100	100
1	B	393/389 (101%)	384 (98%)	9 (2%)	0	100	100
2	C	2/6 (33%)	2 (100%)	0	0	100	100
2	D	2/6 (33%)	2 (100%)	0	0	100	100
All	All	780/790 (99%)	764 (98%)	16 (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	315/325 (97%)	312 (99%)	3 (1%)	76 52
1	B	324/325 (100%)	320 (99%)	4 (1%)	71 45
2	C	2/2 (100%)	2 (100%)	0	100 100
2	D	2/2 (100%)	2 (100%)	0	100 100
All	All	643/654 (98%)	636 (99%)	7 (1%)	81 48

All (7) residues with a non-rotameric sidechain are listed below:

Mol	Chain	Res	Type
1	A	142	HIS
1	A	293[A]	GLN
1	A	293[B]	GLN
1	B	142	HIS
1	B	152	PHE
1	B	258[A]	LYS
1	B	258[B]	LYS

Sometimes sidechains can be flipped to improve hydrogen bonding and reduce clashes. There are no such sidechains identified.

5.3.3 RNA (i)

There are no RNA molecules in this entry.

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

4 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	$\#\|Z\|>2$
2	ALY	C	4	2	10,11,12	0.98	0	7,12,14	0.74	0
2	ALY	D	5	4,2	10,11,12	0.93	0	7,12,14	0.68	0
2	ALY	C	5	4,2	10,11,12	0.91	0	7,12,14	0.58	0
2	ALY	D	4	2	10,11,12	0.90	0	7,12,14	0.81	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
2	ALY	C	4	2	-	0/9/10/12	-
2	ALY	D	5	4,2	-	0/9/10/12	-
2	ALY	C	5	4,2	-	0/9/10/12	-
2	ALY	D	4	2	-	1/9/10/12	-

There are no bond length outliers.

There are no bond angle outliers.

There are no chirality outliers.

All (1) torsion outliers are listed below:

Mol	Chain	Res	Type	Atoms
2	D	4	ALY	CA-CB-CG-CD

There are no ring outliers.

No monomer is involved in short contacts.

5.5 Carbohydrates (i)

There are no monosaccharides in this entry.

5.6 Ligand geometry (i)

Of 7 ligands modelled in this entry, 6 are monoatomic - leaving 1 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	$\#\|Z\| > 2$
5	GOL	B	404	-	5,5,5	0.34	0	5,5,5	0.25	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
5	GOL	B	404	-	-	0/4/4/4	-

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.

No monomer is involved in short contacts.

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	364/389 (93%)	-0.22	4 (1%) 80 79	9, 18, 33, 57	0
1	B	364/389 (93%)	-0.19	9 (2%) 57 56	9, 17, 32, 54	1 (0%)
2	C	2/6 (33%)	0.22	0 100 100	21, 21, 21, 49	0
2	D	2/6 (33%)	-0.15	0 100 100	16, 16, 16, 34	0
All	All	732/790 (92%)	-0.21	13 (1%) 68 68	9, 17, 33, 57	1 (0%)

All (13) RSRZ outliers are listed below:

Mol	Chain	Res	Type	RSRZ
1	A	377	VAL	7.3
1	B	377	VAL	7.1
1	B	14	LEU	3.9
1	B	368	TYR	3.0
1	B	376	VAL	2.6
1	A	253[A]	GLN	2.4
1	A	130	MET	2.4
1	A	375	HIS	2.3
1	B	375	HIS	2.3
1	B	86	GLY	2.2
1	B	253	GLN	2.2
1	B	366	LEU	2.1
1	B	371	GLY	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, 95^{th} 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
2	ALY	C	4	12/13	0.95	0.14	12,28,52,54	0
2	ALY	D	4	12/13	0.97	0.09	10,16,20,21	0
2	ALY	C	5	12/13	0.97	0.12	9,10,12,13	0
2	ALY	D	5	12/13	0.97	0.13	7,8,10,11	0

6.3 Carbohydrates (i)

There are no monosaccharides in this entry.

6.4 Ligands (i)

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, 95^{th} 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
5	GOL	B	404	6/6	0.95	0.08	24,40,46,47	0
3	K	A	402	1/1	0.99	0.04	14,14,14,14	0
3	K	B	401	1/1	1.00	0.07	9,9,9,9	0
3	K	B	402	1/1	1.00	0.06	12,12,12,12	0
4	ZN	A	403	1/1	1.00	0.05	11,11,11,11	0
4	ZN	B	403	1/1	1.00	0.05	10,10,10,10	0
3	K	A	401	1/1	1.00	0.08	10,10,10,10	0

6.5 Other polymers (i)

There are no such residues in this entry.