



wwPDB X-ray Structure Validation Summary Report ⓘ

Oct 23, 2017 – 11:52 AM EDT

PDB ID : 3K1A
Title : Insights into substrate binding at FeMo-cofactor in nitrogenase from the structure of an alpha-70Ile MoFe protein variant
Authors : Peters, J.W.; Sarma, R.; Barney, B.M.; Keable, S.; Seefeldt, L.C.; Dean, D.R.
Deposited on : unknown
Resolution : 2.23 Å(reported)

This is a wwPDB X-ray Structure Validation Summary Report for a publicly released PDB entry.

We welcome your comments at validation@mail.wwpdb.org

A user guide is available at

<http://wwpdb.org/validation/2016/XrayValidationReportHelp>

with specific help available everywhere you see the ⓘ symbol.

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

MolProbity : 4.02b-467
Mogul : 1.7.2 (RC1), CSD as538be (2017)
Xtriage (Phenix) : 1.9-1692
EDS : rb-20030345
Percentile statistics : 20161228.v01 (using entries in the PDB archive December 28th 2016)
Refmac : 5.8.0135
CCP4 : 6.5.0
Ideal geometry (proteins) : Engh & Huber (2001)
Ideal geometry (DNA, RNA) : Parkinson et al. (1996)
Validation Pipeline (wwPDB-VP) : rb-20030345

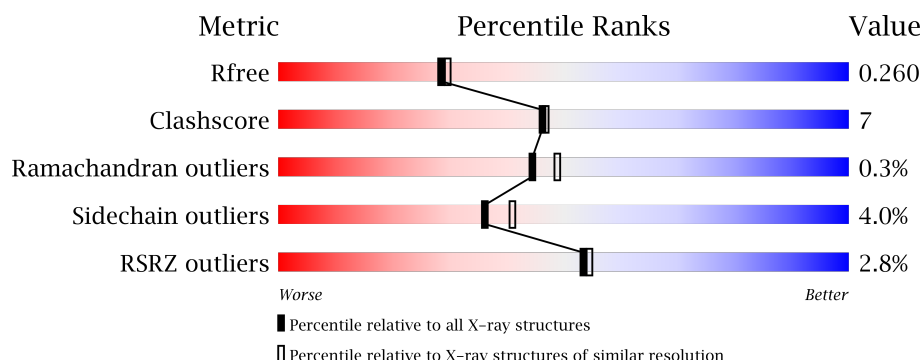
1 Overall quality at a glance

The following experimental techniques were used to determine the structure:

X-RAY DIFFRACTION

The reported resolution of this entry is 2.23 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}	100719	1804 (2.26-2.22)
Clashscore	112137	1957 (2.26-2.22)
Ramachandran outliers	110173	1916 (2.26-2.22)
Sidechain outliers	110143	1917 (2.26-2.22)
RSRZ outliers	101464	1809 (2.26-2.22)

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 ≥ 3 , 2, 1 and 0 types of geometric quality criteria. 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	491	<div> <div>2%</div> <div> <div></div> <div>77%</div> <div>16%</div> <div>5%</div> </div> </div>
1	C	491	<div> <div>4%</div> <div> <div></div> <div>77%</div> <div>16%</div> <div>5%</div> </div> </div>
2	B	522	<div> <div>2%</div> <div> <div></div> <div>83%</div> <div>15%</div> <div></div> </div> </div>
2	D	522	<div> <div>2%</div> <div> <div></div> <div>85%</div> <div>15%</div> <div></div> </div> </div>

The following table lists non-polymeric compounds, carbohydrate monomers and non-standard residues in protein, DNA, RNA chains that are outliers for geometric or electron-density-fit crite-

ria:

Mol	Type	Chain	Res	Chirality	Geometry	Clashes	Electron density
5	CLF	B	6498	-	-	-	X
5	CLF	D	7498	-	-	-	X

2 Entry composition

There are 7 unique types of molecules in this entry. The entry contains 16704 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 Nitrogenase molybdenum-iron protein alpha chain.

Mol	Chain	Residues	Atoms					ZeroOcc	AltConf	Trace
1	A	467	Total	C	N	O	S	0	0	0
			3716	2365	633	694	24			
1	C	468	Total	C	N	O	S	0	0	0
			3714	2365	631	694	24			

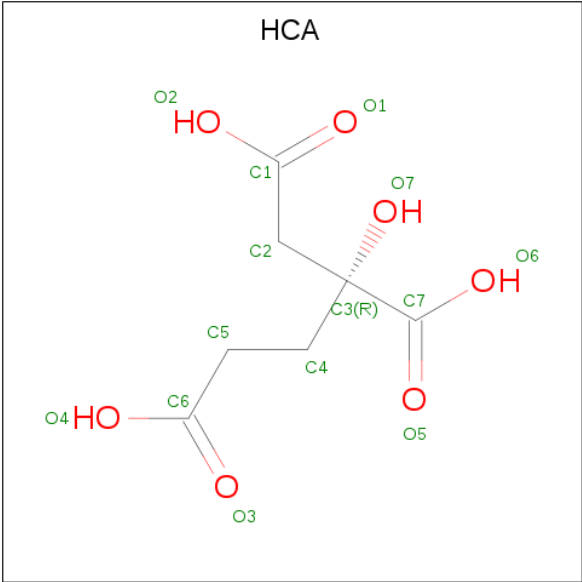
There are 2 discrepancies between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	Comment	Reference
A	70	ILE	VAL	ENGINEERED	UNP P07328
C	70	ILE	VAL	ENGINEERED	UNP P07328

- Molecule 2 is a protein called Nitrogenase molybdenum-iron protein beta chain.

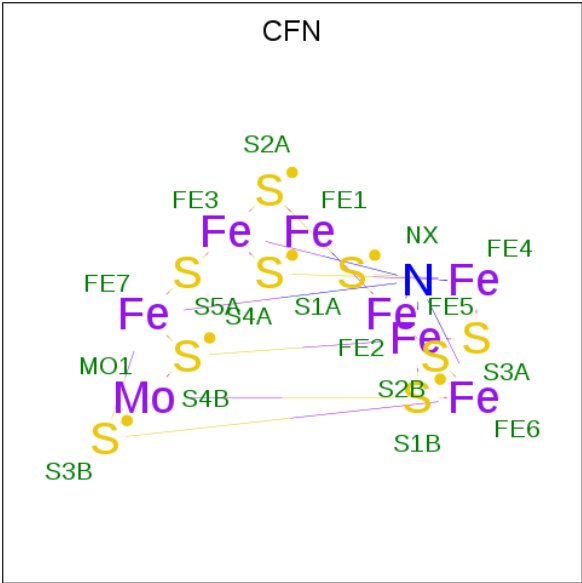
Mol	Chain	Residues	Atoms					ZeroOcc	AltConf	Trace
2	B	522	Total	C	N	O	S	0	0	0
			4174	2666	705	775	28			
2	D	522	Total	C	N	O	S	0	0	0
			4174	2666	705	775	28			

- Molecule 3 is 3-HYDROXY-3-CARBOXY-ADIPIC ACID (three-letter code: HCA) (formula: C₇H₁₀O₇).



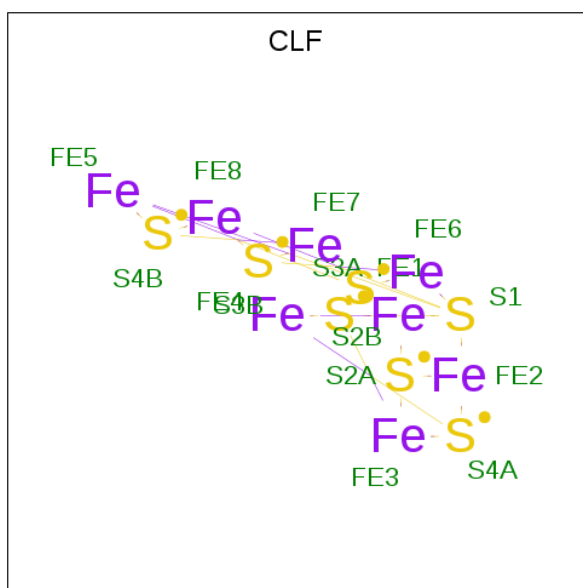
Mol	Chain	Residues	Atoms			ZeroOcc	AltConf
3	A	1	Total	C	O	0	0
			14	7	7		
3	C	1	Total	C	O	0	0
			14	7	7		

- Molecule 4 is FE(7)-MO-S(9)-N CLUSTER (three-letter code: CFN) (formula: Fe₇MoNS₉).



Mol	Chain	Residues	Atoms					ZeroOcc	AltConf
4	A	1	Total 18	Fe 7	Mo 1	N 1	S 9	0	0
4	C	1	Total 18	Fe 7	Mo 1	N 1	S 9	0	0

- Molecule 5 is FE(8)-S(7) CLUSTER (three-letter code: CLF) (formula: Fe₈S₇).



Mol	Chain	Residues	Atoms			ZeroOcc	AltConf
5	B	1	Total	Fe	S	0	0
			15	8	7		
5	D	1	Total	Fe	S	0	0
			15	8	7		

- Molecule 6 is CALCIUM ION (three-letter code: CA) (formula: Ca).

Mol	Chain	Residues	Atoms		ZeroOcc	AltConf
6	B	1	Total	Ca	0	0
			1	1		
6	D	1	Total	Ca	0	0
			1	1		

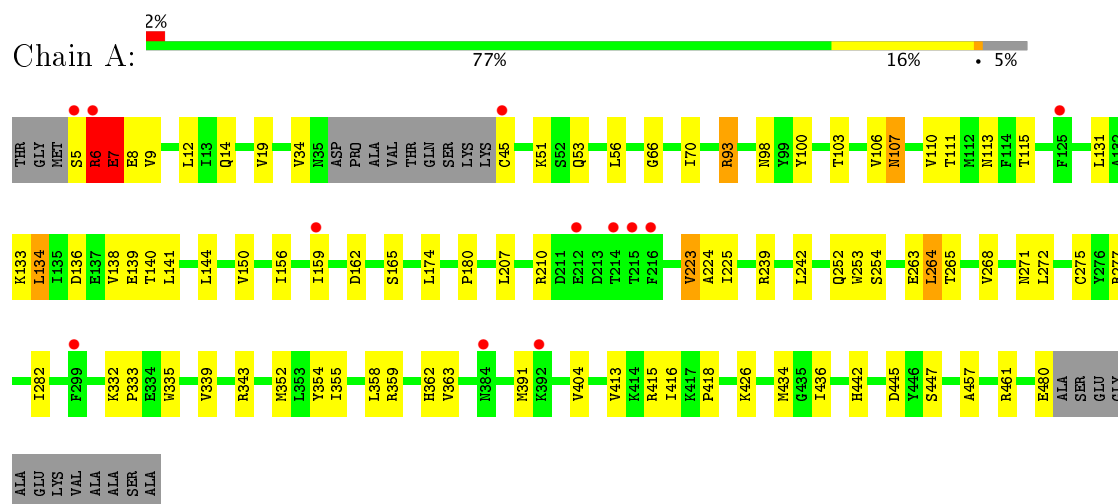
- Molecule 7 is water.

Mol	Chain	Residues	Atoms		ZeroOcc	AltConf
7	A	166	Total	O	0	0
			166	166		
7	B	255	Total	O	0	0
			255	255		
7	C	175	Total	O	0	0
			175	175		
7	D	234	Total	O	0	0
			234	234		

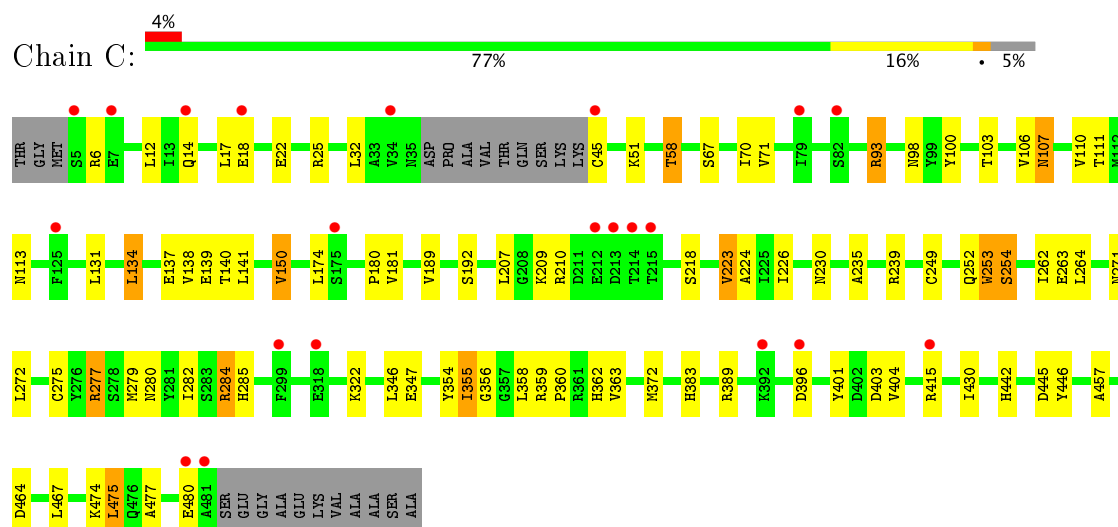
3 Residue-property plots

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

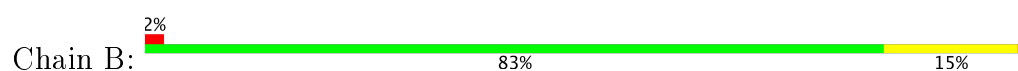
- Molecule 1: Nitrogenase molybdenum-iron protein alpha chain

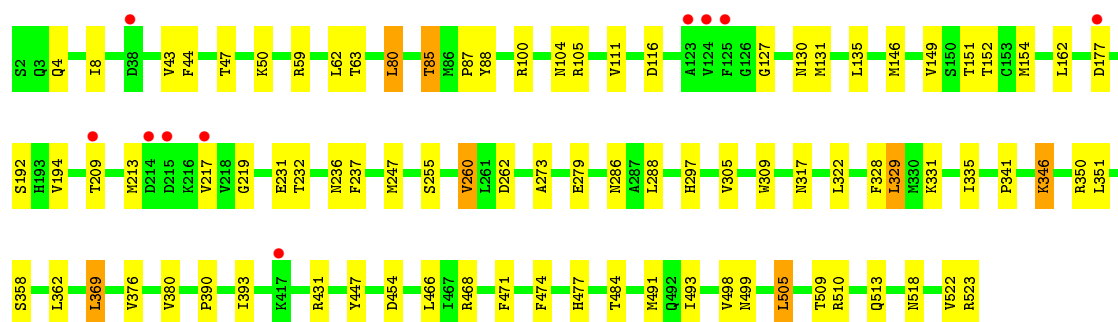


- Molecule 1: Nitrogenase molybdenum-iron protein alpha chain

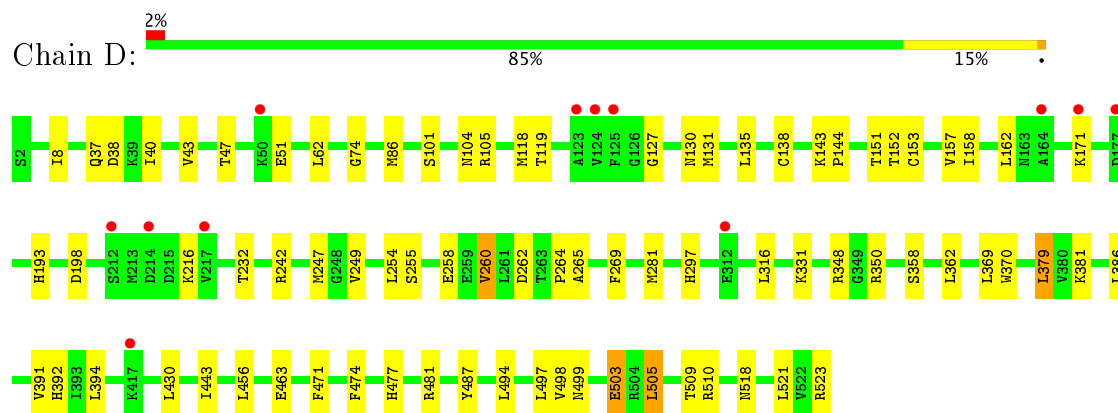


- Molecule 2: Nitrogenase molybdenum-iron protein beta chain





• Molecule 2: Nitrogenase molybdenum-iron protein beta chain



4 Data and refinement statistics

Property	Value	Source
Space group	P 1 21 1	Depositor
Cell constants a, b, c, α , β , γ	77.02Å 129.46Å 107.09Å 90.00° 109.01° 90.00°	Depositor
Resolution (Å)	50.00 – 2.23 27.15 – 2.23	Depositor EDS
% Data completeness (in resolution range)	98.5 (50.00-2.23) 98.5 (27.15-2.23)	Depositor EDS
R_{merge}	(Not available)	Depositor
R_{sym}	0.12	Depositor
$\langle I/\sigma(I) \rangle$ ¹	1.40 (at 2.22Å)	Xtriage
Refinement program	REFMAC 5.2.0019	Depositor
R, R_{free}	0.204 , 0.254 0.209 , 0.260	Depositor DCC
R_{free} test set	4776 reflections (5.27%)	DCC
Wilson B-factor (Å ²)	25.4	Xtriage
Anisotropy	0.141	Xtriage
Bulk solvent k_{sol} (e/Å ³), B_{sol} (Å ²)	0.33 , 30.4	EDS
L-test for twinning ²	$\langle L \rangle = 0.49$, $\langle L^2 \rangle = 0.33$	Xtriage
Estimated twinning fraction	0.018 for h,-k,-h-l	Xtriage
F_o, F_c correlation	0.93	EDS
Total number of atoms	16704	wwPDB-VP
Average B, all atoms (Å ²)	37.0	wwPDB-VP

Xtriage's analysis on translational NCS is as follows: *The largest off-origin peak in the Patterson function is 10.65% 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](#)

Bond lengths and bond angles in the following residue types are not validated in this section: CFN, CLF, HCA, CA

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	0/3802	0.55	0/5125
1	C	0.34	0/3800	0.56	0/5124
2	B	0.33	0/4280	0.53	0/5786
2	D	0.33	0/4280	0.52	0/5786
All	All	0.33	0/16162	0.54	0/21821

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	2
1	C	0	1
All	All	0	3

There are no bond length outliers.

There are no bond angle outliers.

There are no chirality outliers.

All (3) planarity outliers are listed below:

Mol	Chain	Res	Type	Group
1	A	5	SER	Peptide
1	A	6	ARG	Peptide
1	C	253	TRP	Peptide

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	3716	0	3650	59	0
1	C	3714	0	3641	60	0
2	B	4174	0	4089	73	0
2	D	4174	0	4089	59	0
3	A	14	0	6	0	0
3	C	14	0	6	0	0
4	A	18	0	0	2	0
4	C	18	0	0	3	0
5	B	15	0	0	1	0
5	D	15	0	0	2	0
6	B	1	0	0	0	0
6	D	1	0	0	0	0
7	A	166	0	0	4	0
7	B	255	0	0	11	0
7	C	175	0	0	6	0
7	D	234	0	0	3	0
All	All	16704	0	15481	225	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 7.

The worst 5 of 225 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)
2:B:447:TYR:CE1	7:B:779:HOH:O	1.82	1.33
7:B:779:HOH:O	2:D:521:LEU:HD21	1.14	1.25
7:B:779:HOH:O	2:D:521:LEU:CD2	1.81	0.96
2:D:499:ASN:O	2:D:503:GLU:HG3	1.73	0.88
1:C:239:ARG:HE	1:C:252:GLN:HE21	1.19	0.86

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	463/491 (94%)	441 (95%)	20 (4%)	2 (0%)	38	40
1	C	464/491 (94%)	440 (95%)	23 (5%)	1 (0%)	51	58
2	B	520/522 (100%)	509 (98%)	10 (2%)	1 (0%)	51	58
2	D	520/522 (100%)	508 (98%)	11 (2%)	1 (0%)	51	58
All	All	1967/2026 (97%)	1898 (96%)	64 (3%)	5 (0%)	44	49

All (5) Ramachandran outliers are listed below:

Mol	Chain	Res	Type
1	A	6	ARG
1	A	7	GLU
2	B	255	SER
2	D	255	SER
1	C	355	ILE

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	398/414 (96%)	378 (95%)	20 (5%)	28	29
1	C	396/414 (96%)	369 (93%)	27 (7%)	18	16
2	B	454/454 (100%)	444 (98%)	10 (2%)	57	66
2	D	454/454 (100%)	443 (98%)	11 (2%)	54	63
All	All	1702/1736 (98%)	1634 (96%)	68 (4%)	36	41

5 of 68 residues with a non-rotameric sidechain are listed below:

Mol	Chain	Res	Type
1	C	12	LEU
1	C	131	LEU
2	D	260	VAL
1	C	14	GLN
1	C	58	THR

Some sidechains can be flipped to improve hydrogen bonding and reduce clashes. 5 of 43 such sidechains are listed below:

Mol	Chain	Res	Type
2	B	499	ASN
1	C	252	GLN
2	D	499	ASN
2	B	518	ASN
2	B	519	HIS

5.3.3 RNA ⓘ

There are no RNA molecules in this entry.

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

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

5.5 Carbohydrates ⓘ

There are no carbohydrates in this entry.

5.6 Ligand geometry ⓘ

Of 8 ligands modelled in this entry, 2 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	HCA	A	494	-	4,13,13	0.86	0	4,18,18	0.54	0
4	CFN	A	6496	1	18,30,30	8.44	12 (66%)	0,78,78	0.00	-
5	CLF	B	6498	1,2	0,24,24	0.00	-	0,57,57	0.00	-
3	HCA	C	494	-	4,13,13	0.97	0	4,18,18	1.07	0
4	CFN	C	7496	1	18,30,30	8.38	12 (66%)	0,78,78	0.00	-
5	CLF	D	7498	1,2	0,24,24	0.00	-	0,57,57	0.00	-

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	HCA	A	494	-	-	0/7/17/17	0/0/0/0
4	CFN	A	6496	1	-	0/0/204/204	0/0/13/13
5	CLF	B	6498	1,2	-	0/0/132/132	0/12/10/10
3	HCA	C	494	-	-	0/7/17/17	0/0/0/0
4	CFN	C	7496	1	-	0/0/204/204	0/0/13/13
5	CLF	D	7498	1,2	-	0/0/132/132	0/12/10/10

The worst 5 of 24 bond length outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(Å)	Ideal(Å)
4	A	6496	CFN	S1A-FE2	-11.45	2.07	2.29
4	C	7496	CFN	S1A-FE2	-11.39	2.07	2.29
4	A	6496	CFN	S1B-FE6	-11.36	2.07	2.29
4	C	7496	CFN	S3B-FE6	-10.90	2.08	2.29
4	C	7496	CFN	S1B-FE5	-10.89	2.08	2.29

There are no bond angle outliers.

There are no chirality outliers.

There are no torsion outliers.

There are no ring outliers.

4 monomers are involved in 8 short contacts:

Mol	Chain	Res	Type	Clashes	Symm-Clashes
4	A	6496	CFN	2	0
5	B	6498	CLF	1	0
4	C	7496	CFN	3	0
5	D	7498	CLF	2	0

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 [i](#)

6.1 Protein, DNA and RNA chains [i](#)

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	467/491 (95%)	0.21	12 (2%) 56 58	25, 37, 53, 73	0
1	C	468/491 (95%)	0.31	21 (4%) 34 34	27, 37, 55, 73	0
2	B	522/522 (100%)	-0.01	10 (1%) 67 68	27, 35, 49, 63	0
2	D	522/522 (100%)	0.06	12 (2%) 61 62	27, 37, 50, 67	0
All	All	1979/2026 (97%)	0.13	55 (2%) 53 54	25, 36, 52, 73	0

The worst 5 of 55 RSRZ outliers are listed below:

Mol	Chain	Res	Type	RSRZ
2	D	125	PHE	5.3
1	A	212	GLU	4.9
2	B	214	ASP	4.8
1	C	212	GLU	4.6
1	C	481	ALA	4.4

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 carbohydrates 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. 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, 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	LLDF	B-factors(\AA^2)	Q<0.9
5	CLF	B	6498	15/15	0.62	0.23	5.08	21,31,40,46	2
5	CLF	D	7498	15/15	0.64	0.22	3.71	20,28,44,54	0
3	HCA	A	494	14/14	0.96	0.13	-0.56	27,30,34,37	0
3	HCA	C	494	14/14	0.97	0.12	-0.84	30,33,36,39	0
4	CFN	C	7496	18/18	0.94	0.09	-1.83	18,25,32,38	0
4	CFN	A	6496	18/18	0.94	0.09	-2.16	22,27,33,42	0
6	CA	D	524	1/1	0.87	0.08	-2.44	61,61,61,61	0
6	CA	B	524	1/1	0.85	0.07	-2.76	57,57,57,57	0

6.5 Other polymers [i](#)

There are no such residues in this entry.