



Full wwPDB X-ray Structure Validation Report ⓘ

Mar 23, 2026 – 12:32 PM UTC

PDB ID : 5BYQ / pdb_00005byq
Title : Semisynthetic [FeFe]-hydrogenase CpI with oxodithiolato-bridged [2Fe] cofactor
Authors : Esselborn, J.; Muraki, N.; Engelbrecht, V.; Hofmann, E.; Kurisu, G.; Happe, T.
Deposited on : 2015-06-10
Resolution : 1.73 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	:	FAILED
Mogul	:	2022.3.0, CSD as543be (2022)
Xtriage (Phenix)	:	2.0
EDS	:	3.0
Buster-report	:	wwPDB partial adaption of 1.1.7 (2018)
Percentile statistics	:	20250101.v01 (using entries in the PDB archive January 1st 2025)
CCP4	:	9.0.010 (Gargrove)
Density-Fitness	:	1.0.12
Ideal geometry (proteins)	:	Engh & Huber (2001)
Ideal geometry (DNA, RNA)	:	Parkinson et al. (1996)
Validation Pipeline (wwPDB-VP)	:	2.49

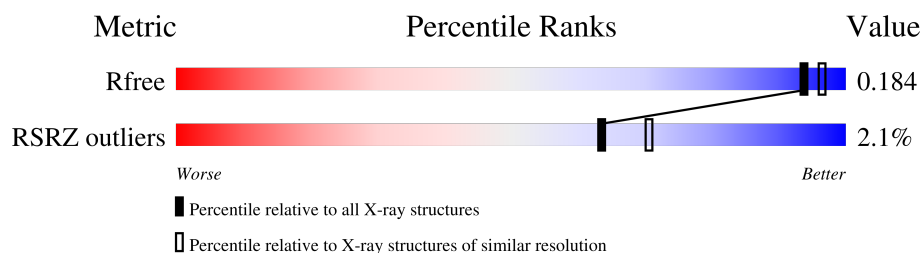
1 Overall quality at a glance

The following experimental techniques were used to determine the structure:

X-RAY DIFFRACTION

The reported resolution of this entry is 1.73 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}	180053	1187 (1.74-1.74)
RSRZ outliers	180081	1188 (1.74-1.74)

MolProbity failed to run properly - the sequence quality summary graphics cannot be shown.

2 Entry composition

There are 7 unique types of molecules in this entry. The entry contains 18886 atoms, of which 8908 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.

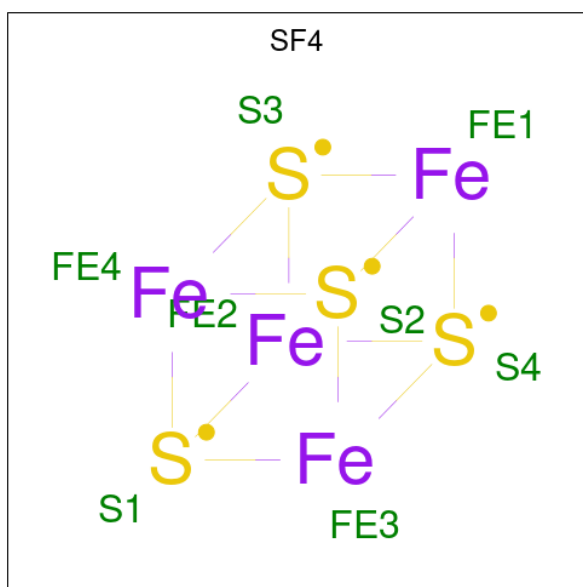
- Molecule 1 is a protein called Iron hydrogenase 1.

Mol	Chain	Residues	Atoms						ZeroOcc	AltConf	Trace
1	A	580	Total	C	H	N	O	S	0	0	0
			8961	2822	4458	776	866	39			
1	B	576	Total	C	H	N	O	S	0	2	0
			8913	2808	4434	769	863	39			

There are 20 discrepancies between the modelled and reference sequences:

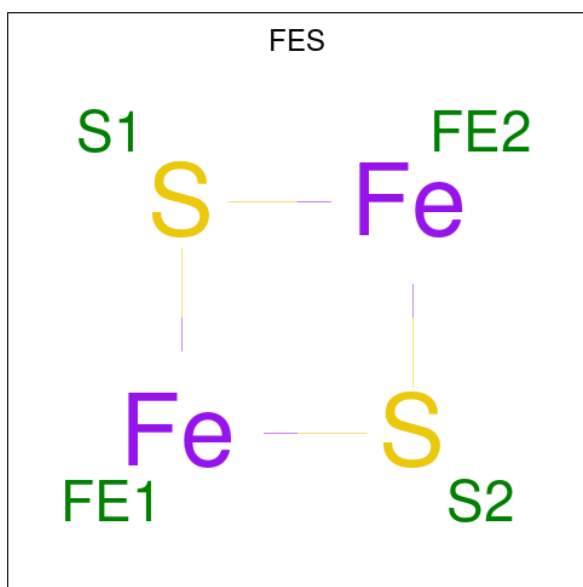
Chain	Residue	Modelled	Actual	Comment	Reference
A	575	SER	-	expression tag	UNP P29166
A	576	ALA	-	expression tag	UNP P29166
A	577	TRP	-	expression tag	UNP P29166
A	578	SER	-	expression tag	UNP P29166
A	579	HIS	-	expression tag	UNP P29166
A	580	PRO	-	expression tag	UNP P29166
A	581	GLN	-	expression tag	UNP P29166
A	582	PHE	-	expression tag	UNP P29166
A	583	GLU	-	expression tag	UNP P29166
A	584	LYS	-	expression tag	UNP P29166
B	575	SER	-	expression tag	UNP P29166
B	576	ALA	-	expression tag	UNP P29166
B	577	TRP	-	expression tag	UNP P29166
B	578	SER	-	expression tag	UNP P29166
B	579	HIS	-	expression tag	UNP P29166
B	580	PRO	-	expression tag	UNP P29166
B	581	GLN	-	expression tag	UNP P29166
B	582	PHE	-	expression tag	UNP P29166
B	583	GLU	-	expression tag	UNP P29166
B	584	LYS	-	expression tag	UNP P29166

- Molecule 2 is IRON/SULFUR CLUSTER (CCD ID: SF4) (formula: Fe₄S₄).



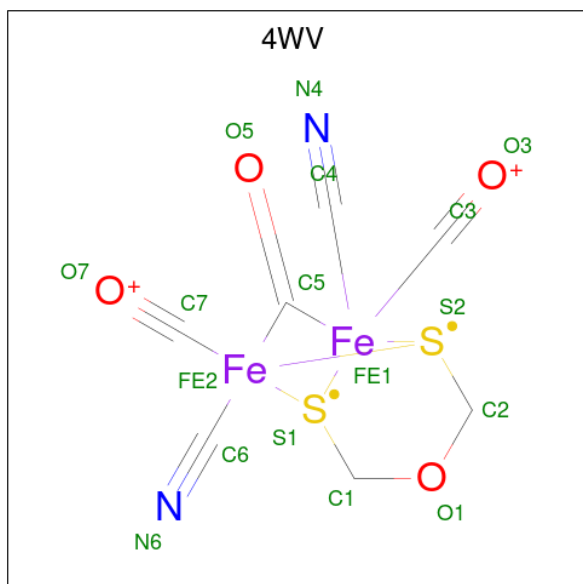
Mol	Chain	Residues	Atoms			ZeroOcc	AltConf
2	A	1	Total	Fe	S	0	0
			8	4	4		
2	A	1	Total	Fe	S	0	0
			8	4	4		
2	A	1	Total	Fe	S	0	0
			8	4	4		
2	A	1	Total	Fe	S	0	0
			8	4	4		
2	B	1	Total	Fe	S	0	0
			8	4	4		
2	B	1	Total	Fe	S	0	0
			8	4	4		
2	B	1	Total	Fe	S	0	0
			8	4	4		

- Molecule 3 is FE2/S2 (INORGANIC) CLUSTER (CCD ID: FES) (formula: Fe_2S_2).



Mol	Chain	Residues	Atoms			ZeroOcc	AltConf
3	A	1	Total	Fe	S	0	0
			4	2	2		
3	B	1	Total	Fe	S	0	0
			4	2	2		

- Molecule 4 is bis(cyano-kappaC)(dicarbonyl)-mu-(oxomethylidene)-mu-(oxydimethanethiolate-1kappaS:2kappaS)diiron(2+) (CCD ID: 4WV) (formula: $C_7H_4Fe_2N_2O_4S_2$).



Mol	Chain	Residues	Atoms						ZeroOcc	AltConf
4	A	1	Total	C	Fe	N	O	S	0	0
			17	7	2	2	4	2		

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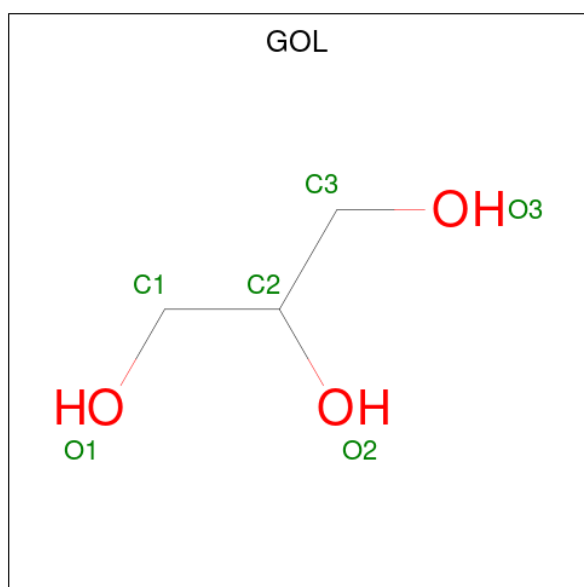
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Mol	Chain	Residues	Atoms					ZeroOcc	AltConf
4	B	1	Total	C	Fe	N	O	S	
			17	7	2	2	4	2	0

- Molecule 5 is MAGNESIUM ION (CCD ID: MG) (formula: Mg).

Mol	Chain	Residues	Atoms		ZeroOcc	AltConf
5	A	2	Total	Mg	0	0
			2	2		
5	B	2	Total	Mg	0	0
			2	2		

- Molecule 6 is GLYCEROL (CCD ID: GOL) (formula: C₃H₈O₃).



Mol	Chain	Residues	Atoms				ZeroOcc	AltConf
6	A	1	Total	C	H	O	0	0
			14	3	8	3		
6	B	1	Total	C	H	O	0	0
			14	3	8	3		

- Molecule 7 is water.

Mol	Chain	Residues	Atoms		ZeroOcc	AltConf
7	A	451	Total	O	0	0
			451	451		
7	B	423	Total	O	0	0
			423	423		

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3 Data and refinement statistics

Property	Value	Source
Space group	P 1 21 1	Depositor
Cell constants a, b, c, α , β , γ	89.66Å 72.45Å 102.94Å 90.00° 96.76° 90.00°	Depositor
Resolution (Å)	47.77 – 1.73 47.77 – 1.73	Depositor EDS
% Data completeness (in resolution range)	100.0 (47.77-1.73) 99.8 (47.77-1.73)	Depositor EDS
R_{merge}	0.08	Depositor
R_{sym}	(Not available)	Depositor
$\langle I/\sigma(I) \rangle$ ¹	1.75 (at 1.73Å)	Xtriage
Refinement program	PHENIX	Depositor
R, R_{free}	0.146 , 0.176 0.157 , 0.184	Depositor DCC
R_{free} test set	1442 reflections (1.05%)	wwPDB-VP
Wilson B-factor (Å ²)	21.3	Xtriage
Anisotropy	0.562	Xtriage
Bulk solvent k_{sol} (e/Å ³), B_{sol} (Å ²)	0.42 , 40.3	EDS
L-test for twinning ²	$\langle L \rangle = 0.49$, $\langle L^2 \rangle = 0.32$	Xtriage
Estimated twinning fraction	No twinning to report.	Xtriage
F_o, F_c correlation	0.97	EDS
Total number of atoms	18886	wwPDB-VP
Average B, all atoms (Å ²)	35.0	wwPDB-VP

Xtriage's analysis on translational NCS is as follows: *The largest off-origin peak in the Patterson function is 6.85% 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.

4 Model quality [i](#)

4.1 Standard geometry [i](#)

MolProbity failed to run properly - this section is therefore empty.

4.2 Too-close contacts [i](#)

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4.3 Torsion angles [i](#)

4.3.1 Protein backbone [i](#)

MolProbity failed to run properly - this section is therefore empty.

4.3.2 Protein sidechains [i](#)

MolProbity failed to run properly - this section is therefore empty.

4.3.3 RNA [i](#)

MolProbity failed to run properly - this section is therefore empty.

4.4 Non-standard residues in protein, DNA, RNA chains [i](#)

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

4.5 Carbohydrates [i](#)

There are no oligosaccharides in this entry.

4.6 Ligand geometry [i](#)

Of 18 ligands modelled in this entry, 4 are monoatomic - leaving 14 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
2	SF4	A	602	1	0,12,12	-	-	-		
3	FES	B	605	1	0,4,4	-	-	-		
2	SF4	A	601	1	0,12,12	-	-	-		
2	SF4	B	604	1	0,12,12	-	-	-		
3	FES	A	605	1	0,4,4	-	-	-		
2	SF4	A	604	1	0,12,12	-	-	-		
2	SF4	A	603	1	0,12,12	-	-	-		
4	4WV	A	606	1	15,19,19	3.79	9 (60%)	3,36,36	11.07	3 (100%)
4	4WV	B	606	1	15,19,19	3.53	7 (46%)	3,36,36	13.01	3 (100%)
6	GOL	B	609	-	5,5,5	0.49	0	5,5,5	0.45	0
2	SF4	B	602	1	0,12,12	-	-	-		
2	SF4	B	601	1	0,12,12	-	-	-		
2	SF4	B	603	1	0,12,12	-	-	-		
6	GOL	A	609	-	5,5,5	0.58	0	5,5,5	0.41	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	SF4	A	602	1	-	-	0/6/5/5
3	FES	B	605	1	-	-	0/1/1/1
2	SF4	A	601	1	-	-	0/6/5/5
2	SF4	B	604	1	-	-	0/6/5/5
3	FES	A	605	1	-	-	0/1/1/1
2	SF4	A	603	1	-	-	0/6/5/5
6	GOL	B	609	-	-	0/4/4/4	-
2	SF4	A	604	1	-	-	0/6/5/5
4	4WV	A	606	1	-	-	0/5/3/3
4	4WV	B	606	1	-	-	0/5/3/3
2	SF4	B	602	1	-	-	0/6/5/5
2	SF4	B	601	1	-	-	0/6/5/5
2	SF4	B	603	1	-	-	0/6/5/5
6	GOL	A	609	-	-	0/4/4/4	-

All (16) bond length outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(Å)	Ideal(Å)
4	B	606	4WV	O1-C2	7.09	1.51	1.41
4	B	606	4WV	C2-S2	6.92	1.90	1.83
4	A	606	4WV	C2-S2	6.58	1.89	1.83
4	A	606	4WV	O1-C2	6.55	1.50	1.41
4	A	606	4WV	S2-FE2	6.17	2.34	2.26
4	A	606	4WV	O1-C1	5.44	1.48	1.41
4	B	606	4WV	O1-C1	5.10	1.48	1.41
4	A	606	4WV	C4-N4	4.19	1.21	1.15
4	B	606	4WV	C4-N4	4.12	1.21	1.15
4	B	606	4WV	S2-FE1	3.83	2.31	2.26
4	A	606	4WV	S2-FE1	3.70	2.31	2.26
4	B	606	4WV	O3-C3	3.04	1.22	1.15
4	A	606	4WV	S1-FE1	3.01	2.30	2.26
4	A	606	4WV	O3-C3	2.74	1.22	1.15
4	A	606	4WV	C1-S1	2.72	1.86	1.83
4	B	606	4WV	S1-FE2	2.13	2.29	2.26

All (6) bond angle outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(°)	Ideal(°)
4	B	606	4WV	O1-C2-S2	-17.40	103.40	115.15
4	A	606	4WV	O1-C1-S1	-14.29	105.50	115.15
4	A	606	4WV	O1-C2-S2	-12.57	106.67	115.15
4	B	606	4WV	O1-C1-S1	-12.28	106.86	115.15
4	B	606	4WV	C2-O1-C1	7.35	121.52	113.07
4	A	606	4WV	C2-O1-C1	-2.36	110.36	113.07

There are no chirality outliers.

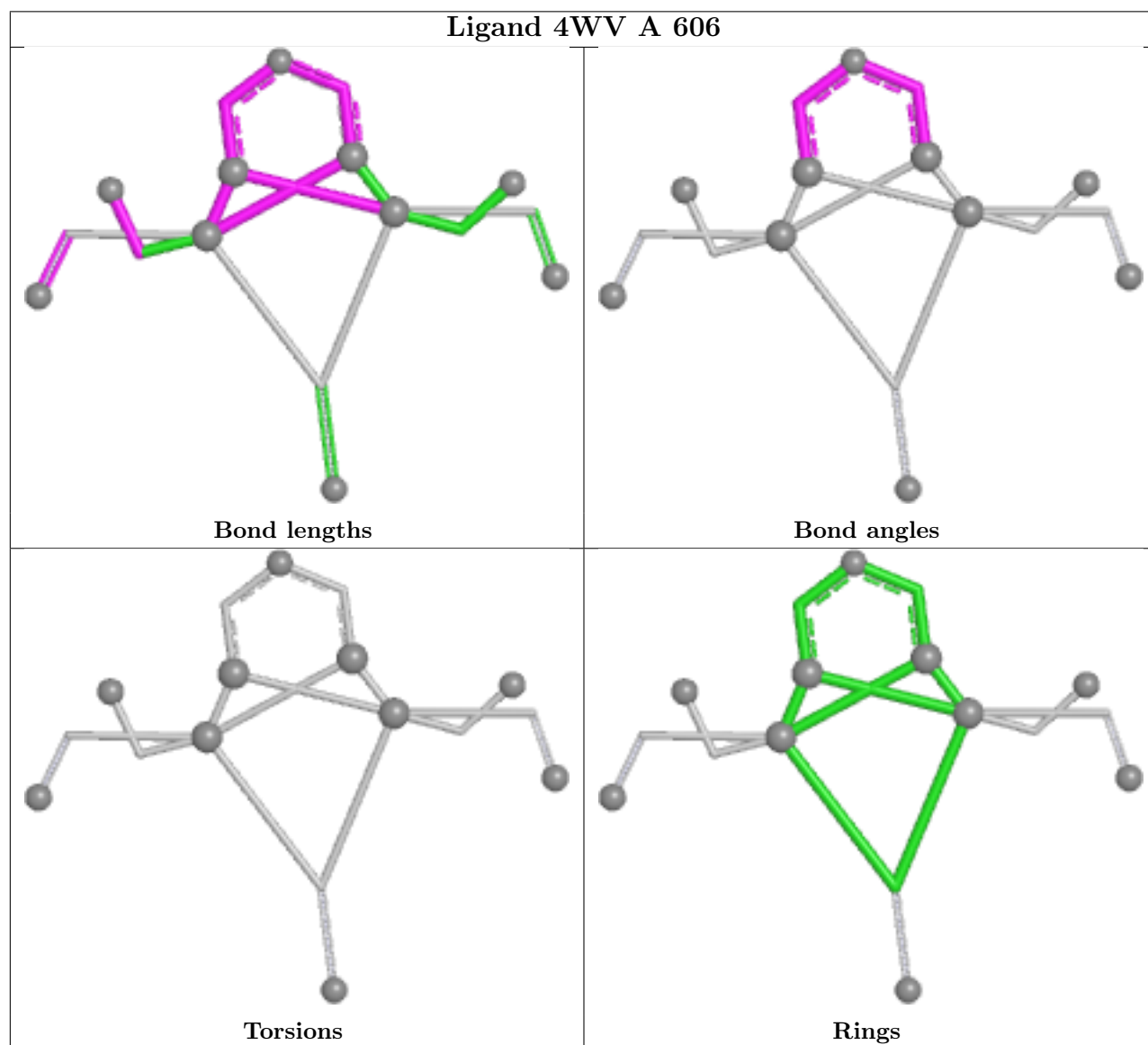
There are no torsion outliers.

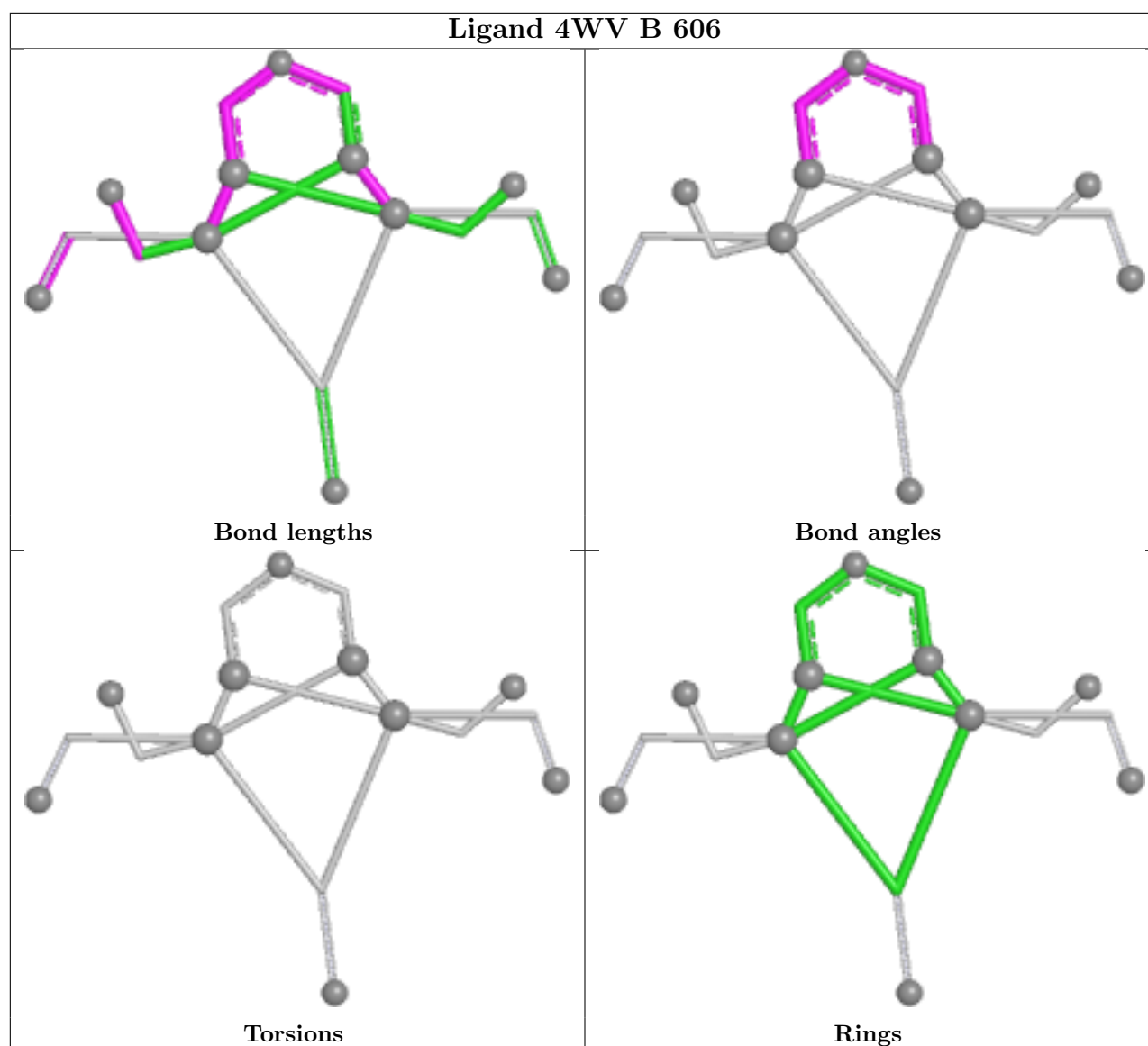
There are no ring outliers.

No monomer is involved in short contacts.

The following is a two-dimensional graphical depiction of Mogul quality analysis of bond lengths, bond angles, torsion angles, and ring geometry for all instances of the Ligand of Interest. In addition, ligands with molecular weight > 250 and outliers as shown on the validation Tables will also be included. For torsion angles, if less than 5% of the Mogul distribution of torsion angles is within 10 degrees of the torsion angle in question, then that torsion angle is considered an outlier. Any bond that is central to one or more torsion angles identified as an outlier by Mogul will be highlighted in the graph. For rings, the root-mean-square deviation (RMSD) between the ring in question and similar rings identified by Mogul is calculated over all ring torsion angles. If the average RMSD is greater than 60 degrees and the minimal RMSD between the ring in question and any Mogul-identified rings is also greater than 60 degrees, then that ring is considered an outlier.

The outliers are highlighted in purple. The color gray indicates Mogul did not find sufficient equivalents in the CSD to analyse the geometry.





4.7 Other polymers [i](#)

There are no such residues in this entry.

4.8 Polymer linkage issues [i](#)

There are no chain breaks in this entry.

5 Fit of model and data [i](#)

5.1 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	580/584 (99%)	-0.15	3 (0%) 87 91	21, 32, 58, 83	0
1	B	576/584 (98%)	-0.16	21 (3%) 46 55	13, 29, 68, 88	2 (0%)
All	All	1156/1168 (98%)	-0.15	24 (2%) 63 70	13, 31, 64, 88	2 (0%)

All (24) RSRZ outliers are listed below:

Mol	Chain	Res	Type	RSRZ
1	B	577	TRP	5.8
1	A	581	GLN	3.5
1	B	576	ALA	3.3
1	B	572	TYR	3.1
1	B	43	ILE	3.1
1	B	69	GLY	3.1
1	B	5	ILE	2.9
1	B	71	ILE	2.9
1	B	575	SER	2.8
1	A	577	TRP	2.7
1	B	2	LYS	2.7
1	B	22	PHE	2.6
1	B	4	ILE	2.5
1	B	6	ILE	2.5
1	B	57	GLY	2.4
1	B	72	ILE	2.4
1	B	11	PHE	2.4
1	B	172	ASN	2.4
1	B	8	GLY	2.2
1	B	3	THR	2.1
1	B	53	VAL	2.1
1	B	9	VAL	2.0
1	A	580	PRO	2.0
1	B	44	ASN	2.0

5.2 Non-standard residues in protein, DNA, RNA chains [i](#)

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

5.3 Carbohydrates [i](#)

There are no oligosaccharides in this entry.

5.4 Ligands [i](#)

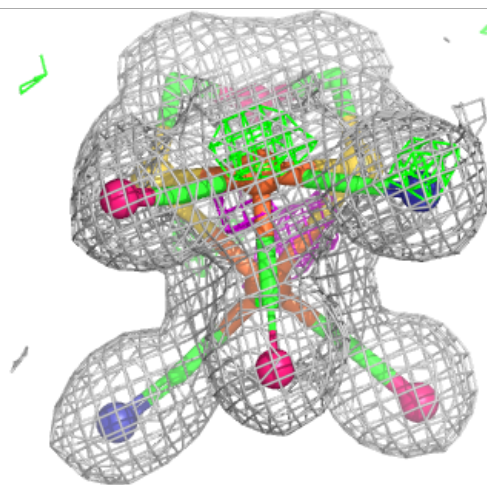
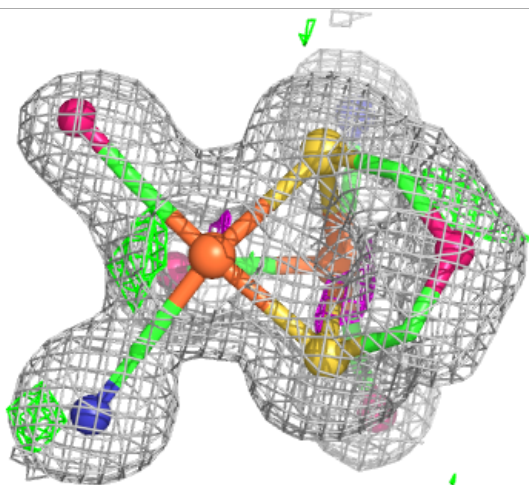
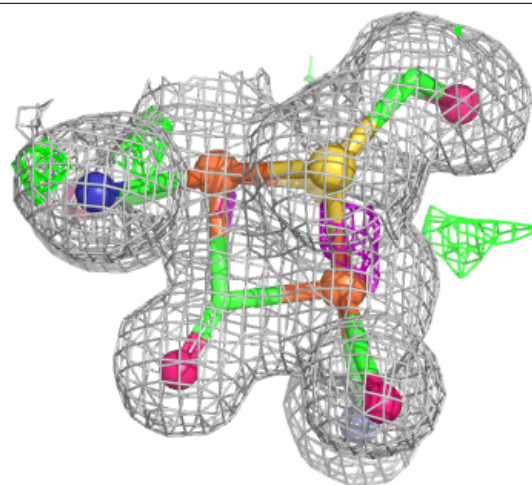
In the following table, the Atoms column lists the number of modelled atoms in the group and the number defined in the chemical component dictionary. The B-factors column lists the minimum, median, 95th percentile and maximum values of B factors of atoms in the group. The column labelled 'Q< 0.9' lists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

Mol	Type	Chain	Res	Atoms	RSCC	RSR	B-factors($\text{\AA}^2$)	Q<0.9
6	GOL	A	609	6/6	0.95	0.08	27,35,44,48	0
6	GOL	B	609	6/6	0.96	0.07	25,33,42,48	0
5	MG	A	607	1/1	0.98	0.03	29,29,29,29	0
5	MG	B	608	1/1	0.98	0.07	33,33,33,33	0
2	SF4	A	604	8/8	0.98	0.04	25,28,29,29	0
2	SF4	B	604	8/8	0.98	0.04	21,25,26,26	0
2	SF4	B	603	8/8	0.99	0.03	18,18,19,20	0
2	SF4	A	602	8/8	0.99	0.03	18,19,20,20	0
3	FES	A	605	4/4	0.99	0.03	21,21,21,22	0
3	FES	B	605	4/4	0.99	0.03	23,24,25,26	0
4	4WV	A	606	17/17	0.99	0.05	17,23,26,26	5
4	4WV	B	606	17/17	0.99	0.04	16,19,23,23	1
2	SF4	A	603	8/8	0.99	0.04	19,20,21,22	0
5	MG	A	608	1/1	0.99	0.05	25,25,25,25	0
2	SF4	A	601	8/8	0.99	0.03	18,19,19,20	0
2	SF4	B	601	8/8	0.99	0.03	15,16,17,17	0
2	SF4	B	602	8/8	0.99	0.03	17,17,18,18	0
5	MG	B	607	1/1	1.00	0.02	29,29,29,29	0

The following is a graphical depiction of the model fit to experimental electron density of all instances of the Ligand of Interest. In addition, ligands with molecular weight > 250 and outliers as shown on the geometry validation Tables will also be included. Each fit is shown from different orientation to approximate a three-dimensional view.

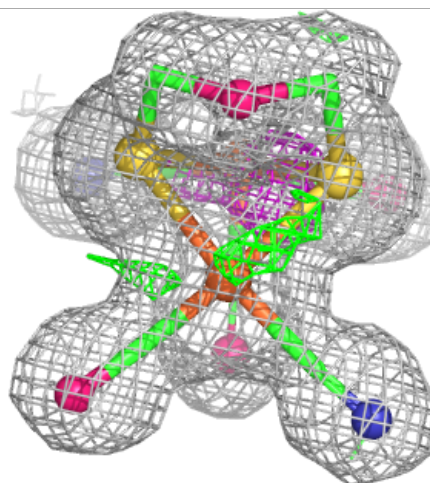
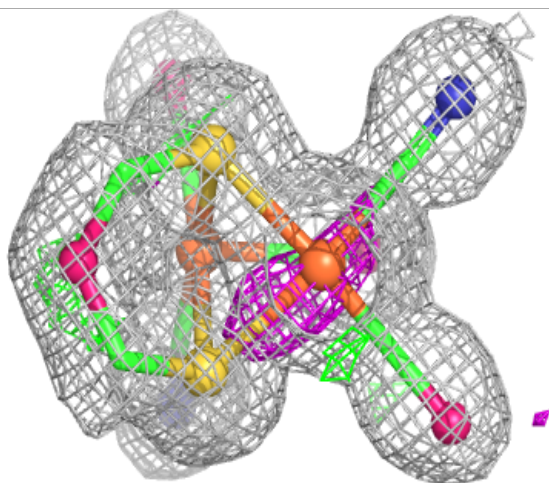
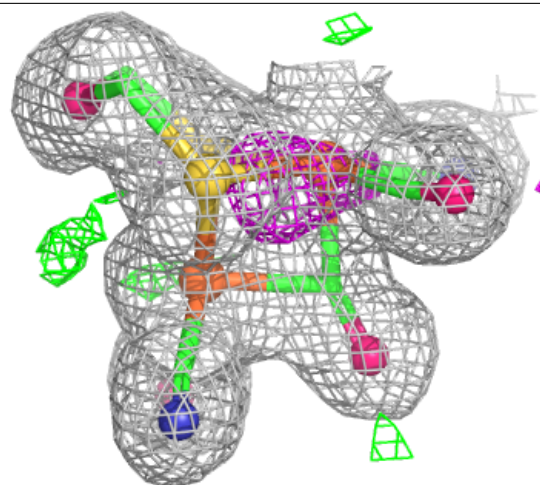
Electron density around 4WV A 606:

$2mF_o-DF_c$ (at 0.7 rmsd) in gray
 mF_o-DF_c (at 3 rmsd) in purple (negative)
and green (positive)



Electron density around 4WV B 606:

$2mF_o-DF_c$ (at 0.7 rmsd) in gray
 mF_o-DF_c (at 3 rmsd) in purple (negative)
and green (positive)



5.5 Other polymers [i](#)

There are no such residues in this entry.