

wwPDB X-ray Structure Validation Summary Report (i)

Feb 26, 2024 – 02:30 PM JST

PDB ID : 8IL4

 $Title \quad : \quad Crystal \ structure \ of \ alcohol \ oxidase \ ParAOX(M59V/Q60P/R61N/F101S/N6)$

02H)(Polyporus arcularius)

Authors: Wu, B.; Wang, Y.

Deposited on : 2023-03-01

Resolution : 3.36 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

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 (1)) 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

buster-report : 1.1.7 (2018)

Percentile statistics : 20191225.v01 (using entries in the PDB archive December 25th 2019)

 $Refmac \quad : \quad 5.8.0158$

CCP4 : 7.0.044 (Gargrove)

Ideal geometry (proteins) : Engh & Huber (2001) Ideal geometry (DNA, RNA) : Parkinson et al. (1996)

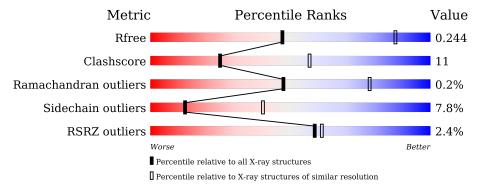
Validation Pipeline (wwPDB-VP) : 2.36

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 3.36 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 $(\# \mathrm{Entries})$	Similar resolution $(\# \text{Entries, resolution range}(\mathring{\mathbf{A}}))$
R_{free}	130704	1558 (3.42-3.30)
Clashscore	141614	1627 (3.42-3.30)
Ramachandran outliers	138981	1599 (3.42-3.30)
Sidechain outliers	138945	1598 (3.42-3.30)
RSRZ outliers	127900	1507 (3.42-3.30)

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 chair	ı	
1	A	649	71%	22%	• 5%
1	В	649	71%	20%	• 6%
1	С	649	68%	25%	• 5%
1	D	649	61%	29%	• 6%



2 Entry composition (i)

There are 2 unique types of molecules in this entry. The entry contains 19024 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 GMC oxidoreductase.

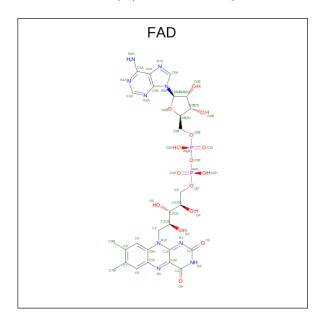
Mol	Chain	Residues	Atoms					ZeroOcc	AltConf	Trace
1	1 A	615	Total	С	N	О	S	0	0	0
1			4765	2994	842	909	20	0	U	
1	В	607	Total	С	N	О	S	0	0	0
1		007	4648	2916	817	896	19	0		
1	C	610	Total	С	N	О	S	0	0	0
1		619	4736	2976	830	910	20	0		
1	D	600	Total	С	N	О	S	0	0	0
1		609	4663	2937	811	895	20	0		

There are 20 discrepancies between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	Comment	Reference
A	59	VAL	MET	engineered mutation	UNP A0A5C3NW19
A	60	PRO	GLN	engineered mutation	UNP A0A5C3NW19
A	61	ASN	ARG	engineered mutation	UNP A0A5C3NW19
A	101	SER	PHE	engineered mutation	UNP A0A5C3NW19
A	602	HIS	ASN	engineered mutation	UNP A0A5C3NW19
В	59	VAL	MET	engineered mutation	UNP A0A5C3NW19
В	60	PRO	GLN	engineered mutation	UNP A0A5C3NW19
В	61	ASN	ARG	engineered mutation	UNP A0A5C3NW19
В	101	SER	PHE	engineered mutation	UNP A0A5C3NW19
В	602	HIS	ASN	engineered mutation	UNP A0A5C3NW19
С	59	VAL	MET	engineered mutation	UNP A0A5C3NW19
С	60	PRO	GLN	engineered mutation	UNP A0A5C3NW19
С	61	ASN	ARG	engineered mutation	UNP A0A5C3NW19
С	101	SER	PHE	engineered mutation	UNP A0A5C3NW19
С	602	HIS	ASN	engineered mutation	UNP A0A5C3NW19
D	59	VAL	MET	engineered mutation	UNP A0A5C3NW19
D	60	PRO	GLN	engineered mutation	UNP A0A5C3NW19
D	61	ASN	ARG	engineered mutation	UNP A0A5C3NW19
D	101	SER	PHE	engineered mutation	UNP A0A5C3NW19
D	602	HIS	ASN	engineered mutation	UNP A0A5C3NW19



 \bullet Molecule 2 is FLAVIN-ADENINE DINUCLEOTIDE (three-letter code: FAD) (formula: $C_{27}H_{33}N_9O_{15}P_2)$ (labeled as "Ligand of Interest" by depositor).



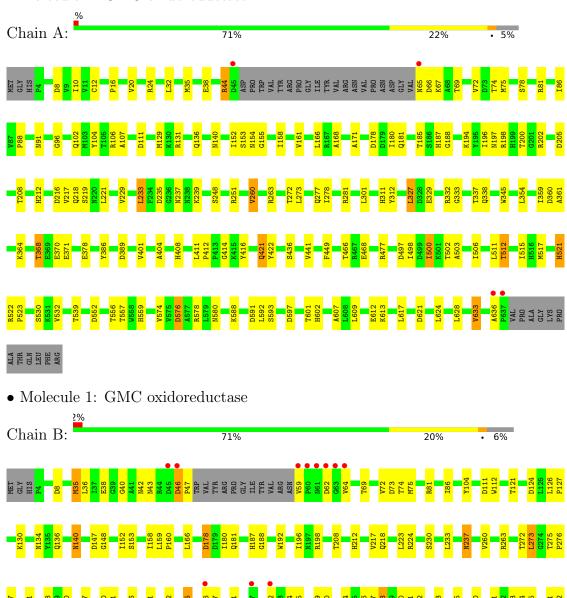
Mol	Chain	Residues	Atoms					ZeroOcc	AltConf	
2	Ω Λ	1	Total	С	N	О	Р	0	0	
2	A	1	53	27	9	15	2	U		
2	В	1	Total	С	N	О	Р	0	0	
2	Ъ	1	53	27	9	15	2	U	U	
2	С	1	Total	С	N	О	Р	0	0	
2		1	53	27	9	15	2	U		
2	9 D) 1	Total	С	N	О	Р	0	0	
	ש	1	53	27	9	15	2	U	U	



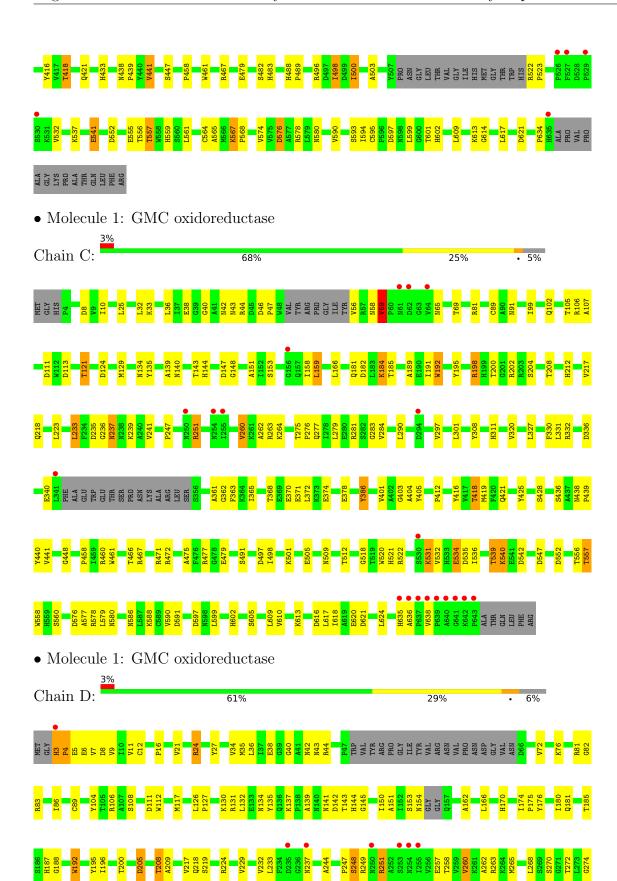
3 Residue-property plots (i)

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.

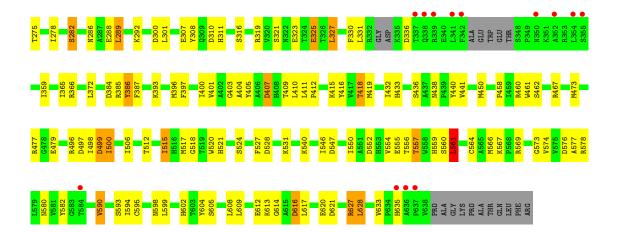
• Molecule 1: GMC oxidoreductase













4 Data and refinement statistics (i)

Property	Value	Source	
Space group	P 1 21 1	Depositor	
Cell constants	71.71Å 187.43Å 118.53Å	Depositor	
a, b, c, α , β , γ	90.00° 90.29° 90.00°	Depositor	
Resolution (Å)	45.57 - 3.36	Depositor	
rtesolution (A)	45.57 - 3.36	EDS	
% Data completeness	94.1 (45.57-3.36)	Depositor	
(in resolution range)	94.1 (45.57-3.36)	EDS	
R_{merge}	(Not available)	Depositor	
R_{sym}	(Not available)	Depositor	
$< I/\sigma(I) > 1$	2.29 (at 3.32Å)	Xtriage	
Refinement program	PHENIX $1.9_1692+SVN$	Depositor	
D D.	0.194 , 0.243	Depositor	
R, R_{free}	0.197 , 0.244	DCC	
R_{free} test set	1996 reflections (4.74%)	wwPDB-VP	
Wilson B-factor (Å ²)	53.5	Xtriage	
Anisotropy	0.430	Xtriage	
Bulk solvent $k_{sol}(e/Å^3)$, $B_{sol}(Å^2)$	0.30 , 17.7	EDS	
L-test for twinning ²	$< L >=0.49, < L^2>=0.32$	Xtriage	
Estimated twinning fraction	0.115 for h,-k,-l	Xtriage	
F_o, F_c correlation	0.91	EDS	
Total number of atoms	19024	wwPDB-VP	
Average B, all atoms (Å ²)	43.0	wwPDB-VP	

Xtriage's analysis on translational NCS is as follows: The largest off-origin peak in the Patterson function is 3.61% of the height of the origin peak. No significant pseudotranslation is detected.

²Theoretical 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.



¹Intensities estimated from amplitudes.

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: FAD

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		
IVIOI		RMSZ	# Z > 5	RMSZ	# Z > 5	
1	A	0.74	0/4880	0.67	0/6630	
1	В	0.71	0/4754	0.65	0/6464	
1	С	0.66	0/4849	0.66	1/6596~(0.0%)	
1	D	0.54	0/4775	0.61	$1/6499 \ (0.0\%)$	
All	All	0.67	0/19258	0.65	$2/26189 \ (0.0\%)$	

There are no bond length outliers.

All (2) bond angle outliers are listed below:

Mol	Chain	Res	Type	Atoms	\mathbf{Z}	$Observed(^o)$	$\operatorname{Ideal}({}^{o})$
1	D	561	LEU	CA-CB-CG	6.37	129.94	115.30
1	С	59	VAL	C-N-CD	5.61	140.18	128.40

There are no chirality outliers.

There are no planarity outliers.

5.2 Too-close contacts (i)

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	4765	0	4608	87	0
1	В	4648	0	4454	84	0
1	С	4736	0	4540	103	0
1	D	4663	0	4436	128	0
2	A	53	0	30	3	0

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Page 10

Mol	Chain	Non-H	H(model)	H(added)	Clashes	Symm-Clashes
2	В	53	0	30	2	0
2	С	53	0	30	2	0
2	D	53	0	30	4	0
All	All	19024	0	18158	402	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 11.

The worst 5 of 402 close contacts within the same asymmetric unit are listed below, sorted by their clash magnitude.

Atom-1	Atom-2	$\begin{array}{c} {\rm Interatomic} \\ {\rm distance} \ ({\rm \AA}) \end{array}$	$egin{aligned} ext{Clash} \ ext{overlap } (ext{Å}) \end{aligned}$	
1:C:184:LYS:HZ3	1:C:185:THR:HG23	1.28	0.95	
1:D:137:LYS:HE3	1:D:154:ASN:HB3	1.55	0.86	
1:D:72:VAL:HG12	1:D:86:ILE:HG12	1.58	0.86	
1:B:136:GLN:O	1:B:198:ARG:NH1	2.08	0.86	
1:D:528:ASP:HB3	1:D:531:LYS:HG3	1.56	0.85	

There are no symmetry-related clashes.

Torsion angles (i) 5.3

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	611/649 (94%)	591 (97%)	20 (3%)	0	100	100	
1	В	601/649 (93%)	573 (95%)	27 (4%)	1 (0%)	47	78	
1	С	613/649 (94%)	584 (95%)	26 (4%)	3 (0%)	29	63	
1	D	599/649 (92%)	570 (95%)	27 (4%)	2 (0%)	41	73	
All	All	2424/2596 (93%)	2318 (96%)	100 (4%)	6 (0%)	47	78	

5 of 6 Ramachandran outliers are listed below:



Mol	Chain	Res	Type
1	D	5	GLU
1	С	47	PRO
1	D	4	PRO
1	С	46	ASP
1	С	59	VAL

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	506/544 (93%)	472 (93%)	34 (7%)		16	47
1	В	489/544 (90%)	453 (93%)	36 (7%)		13	42
1	С	498/544 (92%)	459 (92%)	39 (8%)		12	40
1	D	488/544 (90%)	442 (91%)	46 (9%)		8	31
All	All	1981/2176 (91%)	1826 (92%)	155 (8%)		12	40

5 of 155 residues with a non-rotameric sidechain are listed below:

Mol	Chain	Res	Type
1	D	185	THR
1	D	515	ILE
1	D	219	SER
1	D	327	LEU
1	D	590	VAL

Sometimes sidechains can be flipped to improve hydrogen bonding and reduce clashes. All (4) such sidechains are listed below:

Mol	Chain	Res	Type
1	С	65	ASN
1	С	212	HIS
1	С	421	GLN
1	D	311	HIS



5.3.3 RNA (i)

There are no RNA molecules in this entry.

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

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

5.5 Carbohydrates (i)

There are no monosaccharides in this entry.

5.6 Ligand geometry (i)

4 ligands 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	Chain	Res	Link	В	Bond lengths			Bond angles		
MIOI	туре	Chain	rtes	LIIIK	Counts	RMSZ	# Z > 2	Counts	RMSZ	# Z > 2
2	FAD	С	1000	-	53,58,58	3.74	16 (30%)	68,89,89	2.19	13 (19%)
2	FAD	D	1000	_	53,58,58	3.78	17 (32%)	68,89,89	2.27	18 (26%)
2	FAD	В	1000	-	53,58,58	3.84	17 (32%)	68,89,89	2.42	16 (23%)
2	FAD	A	1000	-	53,58,58	3.92	16 (30%)	68,89,89	2.39	18 (26%)

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	FAD	С	1000	-	-	12/30/50/50	0/6/6/6
2	FAD	D	1000	-	-	10/30/50/50	0/6/6/6
2	FAD	В	1000	-	-	7/30/50/50	0/6/6/6
2	FAD	A	1000	-	-	8/30/50/50	0/6/6/6



The worst	5	of θ	36	bond	length	outliers	are	listed	below.
THE WOLDS	\cdot	OI	,,,	DOM	TOTIE UII	Outilities	$\alpha_{\rm L}$	IIDUCU	DCIOW.

Mol	Chain	Res	Type	Atoms	Z	Observed(A)	Ideal(A)
2	A	1000	FAD	C2B-C1B	-17.54	1.27	1.53
2	В	1000	FAD	C2B-C1B	-17.38	1.27	1.53
2	D	1000	FAD	C2B-C1B	-16.75	1.28	1.53
2	С	1000	FAD	C2B-C1B	-16.52	1.28	1.53
2	A	1000	FAD	O4B-C1B	14.61	1.61	1.41

The worst 5 of 65 bond angle outliers are listed below:

Mol	Chain	Res	Type	Atoms	\mathbf{Z}	$\mathbf{Observed}(^o)$	$\operatorname{Ideal}({}^o)$
2	D	1000	FAD	C5A-C6A-N6A	10.38	136.13	120.35
2	С	1000	FAD	C5A-C6A-N6A	10.28	135.98	120.35
2	В	1000	FAD	C5A-C6A-N6A	10.12	135.74	120.35
2	A	1000	FAD	C5A-C6A-N6A	9.71	135.10	120.35
2	С	1000	FAD	N6A-C6A-N1A	-7.03	103.98	118.57

There are no chirality outliers.

5 of 37 torsion outliers are listed below:

Mol	Chain	Res	Type	Atoms
2	A	1000	FAD	C5B-O5B-PA-O1A
2	A	1000	FAD	C5B-O5B-PA-O2A
2	A	1000	FAD	C3B-C4B-C5B-O5B
2	A	1000	FAD	O4'-C4'-C5'-O5'
2	В	1000	FAD	C5B-O5B-PA-O1A

There are no ring outliers.

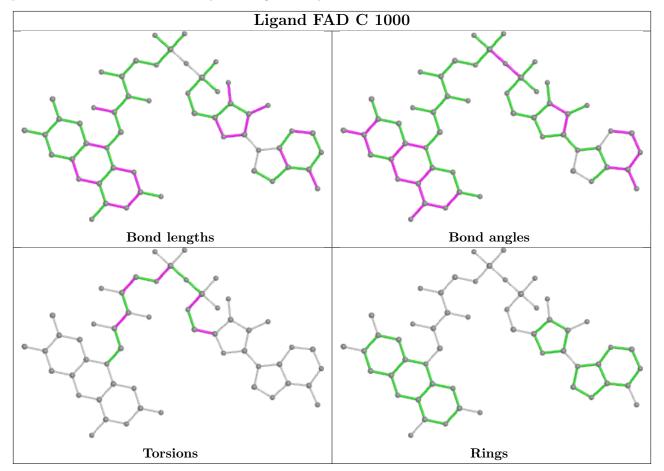
4 monomers are involved in 11 short contacts:

Mol	Chain	Res	Type	Clashes	Symm-Clashes
2	С	1000	FAD	2	0
2	D	1000	FAD	4	0
2	В	1000	FAD	2	0
2	A	1000	FAD	3	0

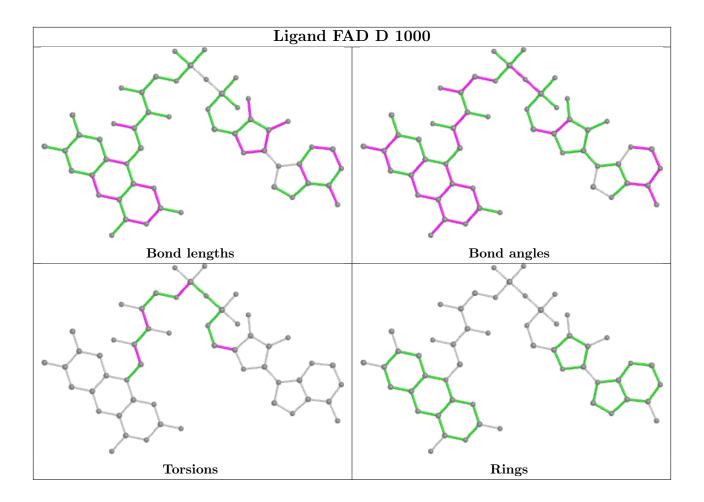
The following is a two-dimensional graphical depiction of Mogul quality analysis of bond lengths, bond angles, torsion angles, and ring geometry for all instances of the Ligand of Interest. In addition, ligands with molecular weight > 250 and outliers as shown on the validation Tables will also be included. For torsion angles, if less then 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



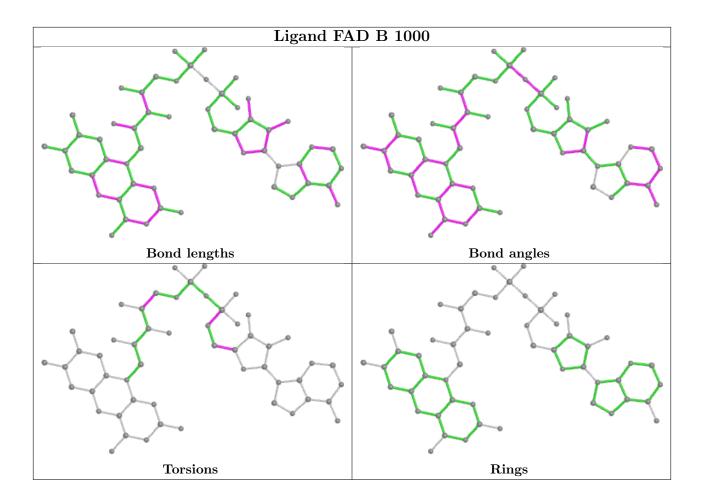
in question and similar rings identified by Mogul is calculated over all ring torsion angles. If the average RMSD is greater than 60 degrees and the minimal RMSD between the ring in question and any Mogul-identified rings is also greater than 60 degrees, then that ring is considered an outlier. The outliers are highlighted in purple. The color gray indicates Mogul did not find sufficient equivalents in the CSD to analyse the geometry.



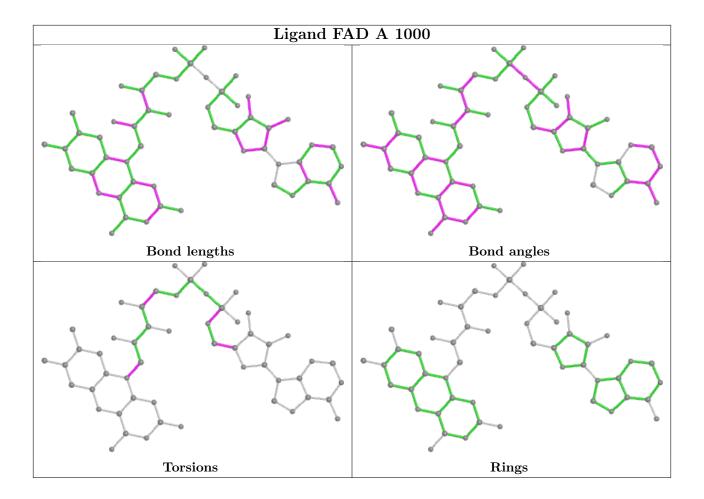












5.7 Other polymers (i)

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, 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>	$\#\mathrm{RSRZ}{>}2$	$\mathbf{OWAB}(\mathbf{\mathring{A}}^2)$	Q < 0.9
1	A	615/649 (94%)	-0.33	4 (0%) 87 91	23, 33, 52, 72	0
1	В	607/649 (93%)	-0.23	16 (2%) 56 58	23, 36, 70, 104	0
1	С	619/649 (95%)	-0.14	19 (3%) 49 52	29, 41, 65, 109	0
1	D	609/649 (93%)	-0.06	20 (3%) 46 48	37, 55, 72, 121	0
All	All	2450/2596 (94%)	-0.19	59 (2%) 59 61	23, 41, 67, 121	0

The worst 5 of 59 RSRZ outliers are listed below:

Mol	Chain	Res	Type	RSRZ
1	A	636	ALA	5.3
1	A	637	PRO	5.2
1	В	62	ASP	4.9
1	С	640	ALA	4.6
1	В	61	ASN	4.5

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 monosaccharides in this entry.

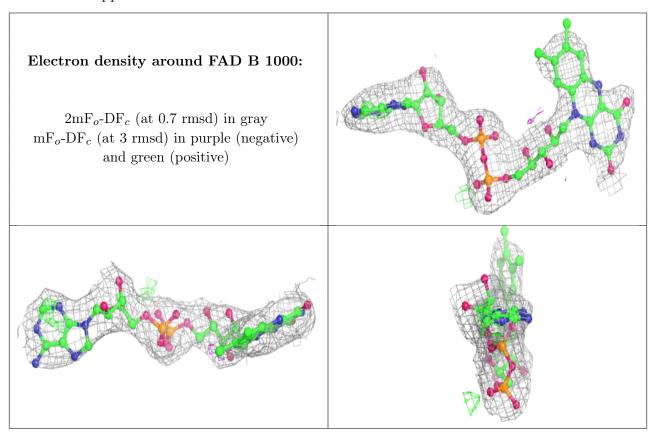
6.4 Ligands (i)

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	${f B-factors}({f \AA}^2)$	Q<0.9
2	FAD	В	1000	53/53	0.95	0.28	58,58,58,58	0
2	FAD	A	1000	53/53	0.96	0.20	31,31,31,31	0
2	FAD	С	1000	53/53	0.96	0.18	25,25,25,25	0
2	FAD	D	1000	53/53	0.96	0.16	31,31,31,31	0

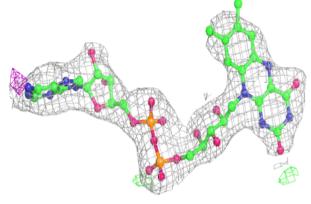
The following is a graphical depiction of the model fit to experimental electron density of all instances of the Ligand of Interest. In addition, ligands with molecular weight > 250 and outliers as shown on the geometry validation Tables will also be included. Each fit is shown from different orientation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

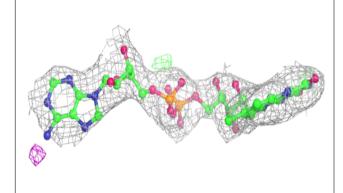


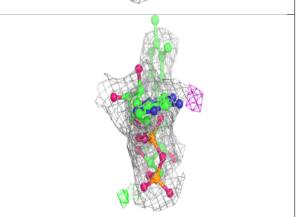


Electron density around FAD A 1000:

 $2 {\rm mF}_o\text{-}{\rm DF}_c$ (at 0.7 rmsd) in gray ${\rm mF}_o\text{-}{\rm DF}_c$ (at 3 rmsd) in purple (negative) and green (positive)

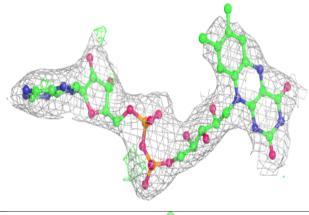


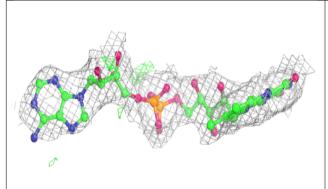


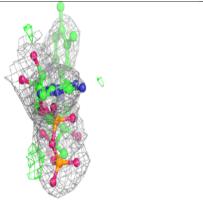


Electron density around FAD C 1000:

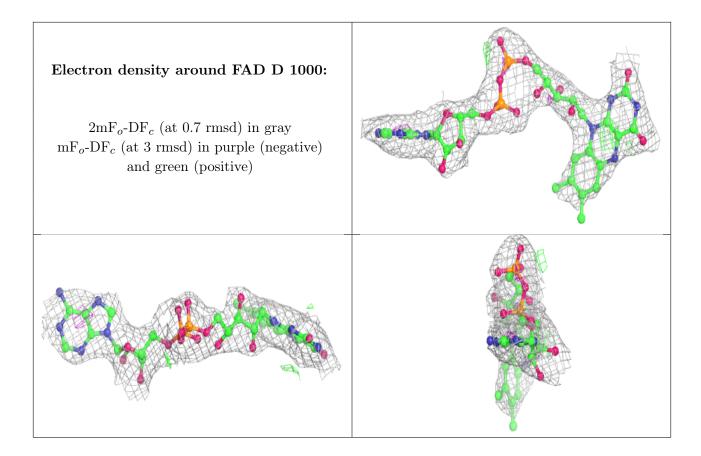
 $2 \mathrm{mF}_o\text{-}\mathrm{DF}_c$ (at 0.7 rmsd) in gray $\mathrm{mF}_o\text{-}\mathrm{DF}_c$ (at 3 rmsd) in purple (negative) and green (positive)











6.5 Other polymers (i)

There are no such residues in this entry.

