



# wwPDB X-ray Structure Validation Summary Report ⓘ

Feb 14, 2017 – 08:44 am GMT

PDB ID : 2FMO  
Title : Ala177Val mutant of E. coli Methylenetetrahydrofolate Reductase  
Authors : Pejchal, R.; Campbell, E.; Guenther, B.D.; Lennon, B.W.; Matthews, R.G.; Ludwig, M.L.  
Deposited on : 2006-01-09  
Resolution : 2.25 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](mailto: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.

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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 : 1.7.2 (RC1), CSD as538be (2017)  
Xtriage (Phenix) : 1.9-1692  
EDS : trunk28620  
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) : recalc28949

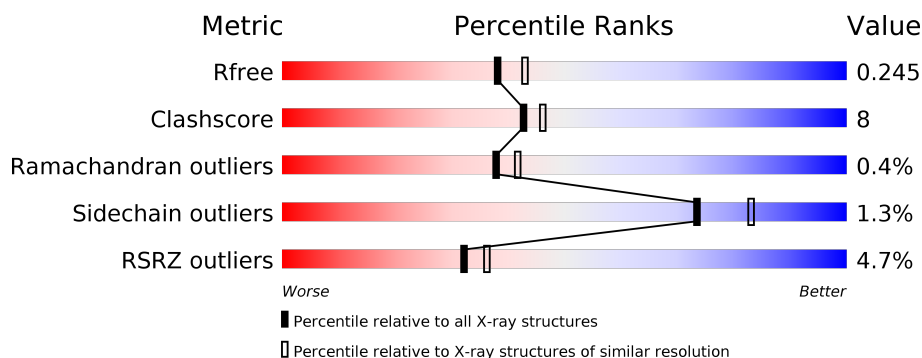
# 1 Overall quality at a glance

The following experimental techniques were used to determine the structure:

*X-RAY DIFFRACTION*

The reported resolution of this entry is 2.25 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	1062 (2.26-2.26)
Clashscore	112137	1178 (2.26-2.26)
Ramachandran outliers	110173	1145 (2.26-2.26)
Sidechain outliers	110143	1146 (2.26-2.26)
RSRZ outliers	101464	1066 (2.26-2.26)

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. 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	304	<div> <div>6%</div> <div> <div></div> <div>72%</div> <div>13%</div> <div>14%</div> </div> </div>
1	B	304	<div> <div>5%</div> <div> <div></div> <div>74%</div> <div>17%</div> <div>9%</div> </div> </div>
1	C	304	<div> <div>2%</div> <div> <div></div> <div>71%</div> <div>15%</div> <div>13%</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 criteria:

Mol	Type	Chain	Res	Chirality	Geometry	Clashes	Electron density
4	MPD	B	896	-	-	-	X

## 2 Entry composition

There are 5 unique types of molecules in this entry. The entry contains 6563 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 5,10-methylenetetrahydrofolate reductase.

Mol	Chain	Residues	Atoms					ZeroOcc	AltConf	Trace
1	A	261	Total	C	N	O	S	0	0	0
			1984	1270	334	369	11			
1	B	276	Total	C	N	O	S	0	0	0
			2093	1330	358	394	11			
1	C	265	Total	C	N	O	S	0	0	0
			2019	1283	345	380	11			

There are 27 discrepancies between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	Comment	Reference
A	177	VAL	ALA	ENGINEERED	UNP P0AEZ1
A	297	LEU	-	CLONING ARTIFACT	UNP P0AEZ1
A	298	GLU	-	CLONING ARTIFACT	UNP P0AEZ1
A	299	HIS	-	EXPRESSION TAG	UNP P0AEZ1
A	300	HIS	-	EXPRESSION TAG	UNP P0AEZ1
A	301	HIS	-	EXPRESSION TAG	UNP P0AEZ1
A	302	HIS	-	EXPRESSION TAG	UNP P0AEZ1
A	303	HIS	-	EXPRESSION TAG	UNP P0AEZ1
A	304	HIS	-	EXPRESSION TAG	UNP P0AEZ1
B	177	VAL	ALA	ENGINEERED	UNP P0AEZ1
B	297	LEU	-	CLONING ARTIFACT	UNP P0AEZ1
B	298	GLU	-	CLONING ARTIFACT	UNP P0AEZ1
B	299	HIS	-	EXPRESSION TAG	UNP P0AEZ1
B	300	HIS	-	EXPRESSION TAG	UNP P0AEZ1
B	301	HIS	-	EXPRESSION TAG	UNP P0AEZ1
B	302	HIS	-	EXPRESSION TAG	UNP P0AEZ1
B	303	HIS	-	EXPRESSION TAG	UNP P0AEZ1
B	304	HIS	-	EXPRESSION TAG	UNP P0AEZ1
C	177	VAL	ALA	ENGINEERED	UNP P0AEZ1
C	297	LEU	-	CLONING ARTIFACT	UNP P0AEZ1
C	298	GLU	-	CLONING ARTIFACT	UNP P0AEZ1
C	299	HIS	-	EXPRESSION TAG	UNP P0AEZ1
C	300	HIS	-	EXPRESSION TAG	UNP P0AEZ1

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Chain	Residue	Modelled	Actual	Comment	Reference
C	301	HIS	-	EXPRESSION TAG	UNP P0AEZ1
C	302	HIS	-	EXPRESSION TAG	UNP P0AEZ1
C	303	HIS	-	EXPRESSION TAG	UNP P0AEZ1
C	304	HIS	-	EXPRESSION TAG	UNP P0AEZ1

- Molecule 2 is SULFATE ION (three-letter code: SO4) (formula: O<sub>4</sub>S).



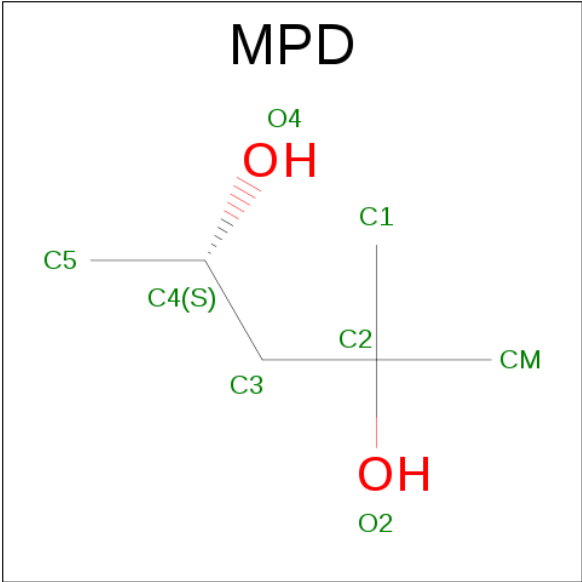
Mol	Chain	Residues	Atoms			ZeroOcc	AltConf
2	A	1	Total	O	S	0	0
			5	4	1		
2	A	1	Total	O	S	0	0
			5	4	1		
2	B	1	Total	O	S	0	0
			5	4	1		
2	B	1	Total	O	S	0	0
			5	4	1		
2	C	1	Total	O	S	0	0
			5	4	1		

- Molecule 3 is FLAVIN-ADENINE DINUCLEOTIDE (three-letter code: FAD) (formula: C<sub>27</sub>H<sub>33</sub>N<sub>9</sub>O<sub>15</sub>P<sub>2</sub>).



Mol	Chain	Residues	Atoms					ZeroOcc	AltConf
3	A	1	Total	C	N	O	P	0	0
			53	27	9	15	2		
3	B	1	Total	C	N	O	P	0	0
			53	27	9	15	2		
3	C	1	Total	C	N	O	P	0	0
			53	27	9	15	2		

- Molecule 4 is (4S)-2-METHYL-2,4-PENTANEDIOL (three-letter code: MPD) (formula: C<sub>6</sub>H<sub>14</sub>O<sub>2</sub>).



Mol	Chain	Residues	Atoms			ZeroOcc	AltConf
4	A	1	Total	C	O	0	0
			8	6	2		
4	B	1	Total	C	O	0	0
			8	6	2		
4	C	1	Total	C	O	0	0
			8	6	2		

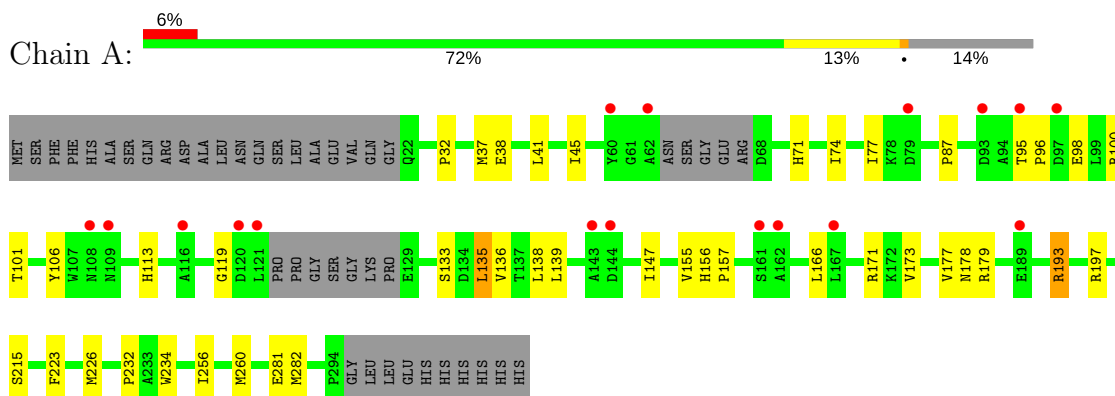
- Molecule 5 is water.

Mol	Chain	Residues	Atoms		ZeroOcc	AltConf
5	A	50	Total	O	0	0
			50	50		
5	B	105	Total	O	0	0
			105	105		
5	C	104	Total	O	0	0
			104	104		

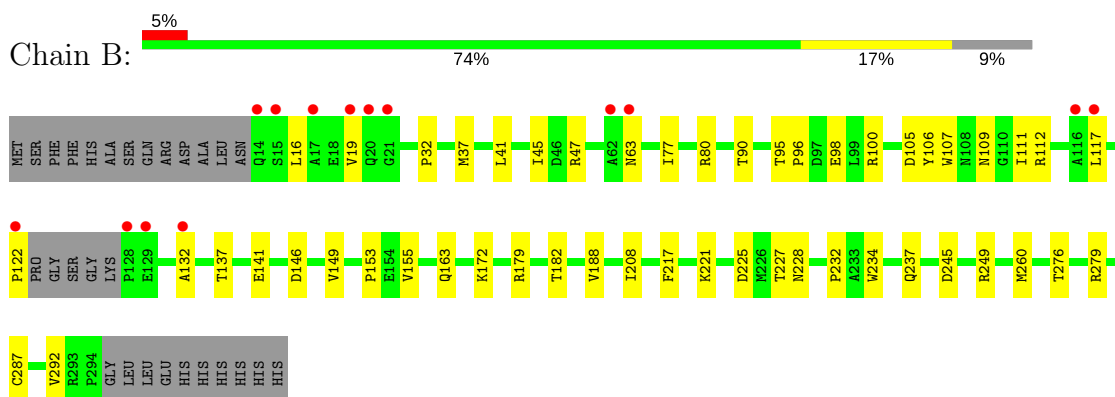
### 3 Residue-property plots [i](#)

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.

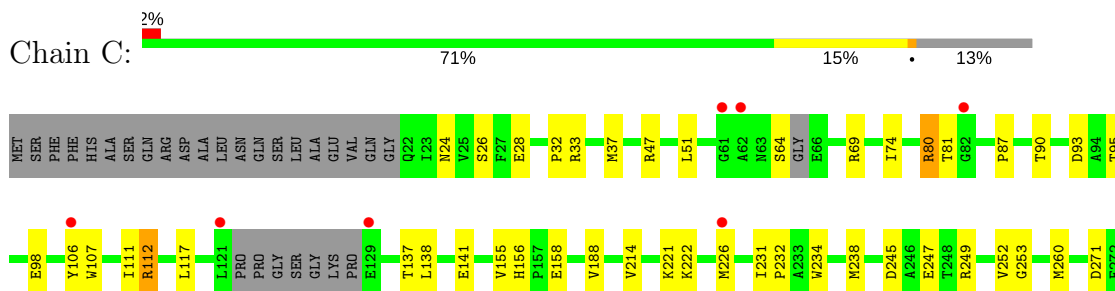
- Molecule 1: 5,10-methylenetetrahydrofolate reductase



- Molecule 1: 5,10-methylenetetrahydrofolate reductase



- Molecule 1: 5,10-methylenetetrahydrofolate reductase





H273		C287		V292	R293	P294	GLY	LEU	LEU	GLU	HIS	HIS	HIS	HIS	HIS	HIS
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## 4 Data and refinement statistics

Property	Value	Source
Space group	C 1 2 1	Depositor
Cell constants a, b, c, $\alpha$ , $\beta$ , $\gamma$	102.15Å 128.35Å 97.58Å 90.00° 121.07° 90.00°	Depositor
Resolution (Å)	20.01 – 2.25 20.01 – 2.10	Depositor EDS
% Data completeness (in resolution range)	99.7 (20.01-2.25) 99.7 (20.01-2.10)	Depositor EDS
$R_{merge}$	(Not available)	Depositor
$R_{sym}$	0.05	Depositor
$\langle I/\sigma(I) \rangle$ <sup>1</sup>	2.15 (at 2.09Å)	Xtriage
Refinement program	CNS 1.1	Depositor
R, $R_{free}$	0.219 , 0.247 0.217 , 0.245	Depositor DCC
$R_{free}$ test set	2583 reflections (5.08%)	DCC
Wilson B-factor (Å <sup>2</sup> )	30.6	Xtriage
Anisotropy	0.081	Xtriage
Bulk solvent $k_{sol}$ (e/Å <sup>3</sup> ), $B_{sol}$ (Å <sup>2</sup> )	0.34 , 55.0	EDS
L-test for twinning <sup>2</sup>	$\langle  L  \rangle = 0.50$ , $\langle L^2 \rangle = 0.33$	Xtriage
Estimated twinning fraction	0.014 for h,-k,-h-l	Xtriage
$F_o, F_c$ correlation	0.93	EDS
Total number of atoms	6563	wwPDB-VP
Average B, all atoms (Å <sup>2</sup> )	32.0	wwPDB-VP

Xtriage's analysis on translational NCS is as follows: *The largest off-origin peak in the Patterson function is 3.78% of the height of the origin peak. No significant pseudotranslation is detected.*

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

<sup>2</sup>Theoretical 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: MPD, SO4, 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	
		RMSZ	$\# Z  > 5$	RMSZ	$\# Z  > 5$
1	A	0.33	0/2025	0.55	0/2755
1	B	0.37	0/2137	0.60	1/2910 (0.0%)
1	C	0.37	0/2060	0.58	0/2802
All	All	0.36	0/6222	0.58	1/8467 (0.0%)

There are no bond length outliers.

All (1) bond angle outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(°)	Ideal(°)
1	B	146	ASP	N-CA-C	-5.37	96.50	111.00

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	1984	0	1901	31	1
1	B	2093	0	1990	32	0
1	C	2019	0	1917	34	0
2	A	10	0	0	0	0
2	B	10	0	0	0	0
2	C	5	0	0	0	0
3	A	53	0	31	1	0

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Mol	Chain	Non-H	H(model)	H(added)	Clashes	Symm-Clashes
3	B	53	0	31	0	0
3	C	53	0	31	0	0
4	A	8	0	13	1	0
4	B	8	0	13	0	0
4	C	8	0	13	0	0
5	A	50	0	0	2	0
5	B	105	0	0	1	0
5	C	104	0	0	1	0
All	All	6563	0	5940	96	1

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 8.

The worst 5 of 96 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:C:80:ARG:HG3	1:C:81:THR:HG23	1.61	0.82
1:A:223:PHE:HA	1:A:226:MET:HE3	1.61	0.80
1:C:188:VAL:HG22	1:C:260:MET:HG3	1.64	0.77
1:C:32:PRO:HB3	1:C:37:MET:HG3	1.70	0.73
1:C:69:ARG:NH1	5:C:509:HOH:O	2.23	0.72

All (1) symmetry-related close contacts are listed below. The label for Atom-2 includes the symmetry operator and encoded unit-cell translations to be applied.

Atom-1	Atom-2	Interatomic distance (Å)	Clash overlap (Å)
1:A:171:ARG:NH1	1:A:171:ARG:NH1[2_455]	1.98	0.22

## 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	255/304 (84%)	246 (96%)	8 (3%)	1 (0%)	38	42
1	B	272/304 (90%)	270 (99%)	1 (0%)	1 (0%)	38	42
1	C	259/304 (85%)	253 (98%)	5 (2%)	1 (0%)	38	42
All	All	786/912 (86%)	769 (98%)	14 (2%)	3 (0%)	38	42

All (3) Ramachandran outliers are listed below:

Mol	Chain	Res	Type
1	B	155	VAL
1	C	155	VAL
1	A	155	VAL

### 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	202/259 (78%)	200 (99%)	2 (1%)	80	87
1	B	213/259 (82%)	210 (99%)	3 (1%)	71	81
1	C	206/259 (80%)	203 (98%)	3 (2%)	70	79
All	All	621/777 (80%)	613 (99%)	8 (1%)	73	83

5 of 8 residues with a non-rotameric sidechain are listed below:

Mol	Chain	Res	Type
1	B	225	ASP
1	C	112	ARG
1	C	80	ARG
1	B	163	GLN
1	B	237	GLN

Some sidechains can be flipped to improve hydrogen bonding and reduce clashes. 5 of 9 such sidechains are listed below:

Mol	Chain	Res	Type
1	B	109	ASN
1	C	273	HIS
1	C	156	HIS
1	A	228	ASN
1	B	163	GLN

### 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 ⓘ

11 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	Res	Link	Bond lengths			Bond angles		
					Counts	RMSZ	# Z  > 2	Counts	RMSZ	# Z  > 2
3	FAD	A	395	-	51,58,58	2.09	10 (19%)	54,89,89	2.96	16 (29%)
4	MPD	A	895	-	7,7,7	1.22	1 (14%)	9,10,10	0.32	0
2	SO4	A	900	-	4,4,4	0.36	0	6,6,6	0.09	0
2	SO4	A	901	-	4,4,4	0.33	0	6,6,6	0.10	0
3	FAD	B	396	-	51,58,58	2.24	16 (31%)	54,89,89	2.94	18 (33%)
4	MPD	B	896	-	7,7,7	1.21	1 (14%)	9,10,10	0.34	0
2	SO4	B	902	-	4,4,4	0.31	0	6,6,6	0.10	0
2	SO4	B	903	-	4,4,4	0.39	0	6,6,6	0.10	0
3	FAD	C	397	-	51,58,58	1.80	12 (23%)	54,89,89	2.82	13 (24%)

Mol	Type	Chain	Res	Link	Bond lengths			Bond angles		
					Counts	RMSZ	# Z  > 2	Counts	RMSZ	# Z  > 2
4	MPD	C	897	-	7,7,7	1.21	1 (14%)	9,10,10	0.31	0
2	SO4	C	904	-	4,4,4	0.31	0	6,6,6	0.15	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
3	FAD	A	395	-	-	0/28/50/50	0/6/6/6
4	MPD	A	895	-	-	0/5/5/5	0/0/0/0
2	SO4	A	900	-	-	0/0/0/0	0/0/0/0
2	SO4	A	901	-	-	0/0/0/0	0/0/0/0
3	FAD	B	396	-	-	0/28/50/50	0/6/6/6
4	MPD	B	896	-	-	0/5/5/5	0/0/0/0
2	SO4	B	902	-	-	0/0/0/0	0/0/0/0
2	SO4	B	903	-	-	0/0/0/0	0/0/0/0
3	FAD	C	397	-	-	0/28/50/50	0/6/6/6
4	MPD	C	897	-	-	0/5/5/5	0/0/0/0
2	SO4	C	904	-	-	0/0/0/0	0/0/0/0

The worst 5 of 41 bond length outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(Å)	Ideal(Å)
3	B	396	FAD	O5B-C5B	-3.36	1.31	1.44
3	B	396	FAD	C8A-N7A	-3.20	1.28	1.34
4	A	895	MPD	O2-C2	-3.13	1.36	1.44
4	B	896	MPD	O2-C2	-3.10	1.36	1.44
4	C	897	MPD	O2-C2	-3.08	1.36	1.44

The worst 5 of 47 bond angle outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(°)	Ideal(°)
3	C	397	FAD	C4X-C10-N10	-7.45	115.35	120.52
3	A	395	FAD	C4X-C10-N10	-6.85	115.76	120.52
3	B	396	FAD	C4X-C10-N10	-6.75	115.83	120.52
3	A	395	FAD	C4X-C4-N3	-5.67	115.40	123.48
3	C	397	FAD	C4X-C4-N3	-5.58	115.53	123.48

There are no chirality outliers.

There are no torsion outliers.

There are no ring outliers.

2 monomers are involved in 2 short contacts:

Mol	Chain	Res	Type	Clashes	Symm-Clashes
3	A	395	FAD	1	0
4	A	895	MPD	1	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, 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	261/304 (85%)	0.40	17 (6%) 20 22	20, 38, 57, 64	0
1	B	276/304 (90%)	0.08	14 (5%) 29 33	15, 28, 48, 63	0
1	C	265/304 (87%)	-0.13	7 (2%) 56 60	16, 28, 42, 53	0
All	All	802/912 (87%)	0.11	38 (4%) 32 36	15, 31, 53, 64	0

The worst 5 of 38 RSRZ outliers are listed below:

Mol	Chain	Res	Type	RSRZ
1	A	121	LEU	6.1
1	C	121	LEU	4.7
1	A	143	ALA	4.1
1	A	120	ASP	4.0
1	A	93	ASP	3.9

### 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, 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	RSCC	RSR	LLDF	B-factors(Å <sup>2</sup> )	Q<0.9
4	MPD	B	896	8/8	0.86	0.21	2.30	39,40,40,41	0
4	MPD	C	897	8/8	0.86	0.18	0.96	37,38,40,41	0
2	SO4	A	901	5/5	0.95	0.15	0.90	53,54,54,55	5
4	MPD	A	895	8/8	0.94	0.19	0.89	40,42,44,44	0
2	SO4	C	904	5/5	0.97	0.13	0.34	43,44,45,45	5
2	SO4	B	902	5/5	0.97	0.12	-0.28	39,41,41,43	5
3	FAD	B	396	53/53	0.93	0.13	-0.39	19,24,39,41	0
3	FAD	A	395	53/53	0.89	0.15	-0.48	26,35,55,56	0
3	FAD	C	397	53/53	0.95	0.10	-0.94	19,22,36,38	0
2	SO4	B	903	5/5	0.94	0.22	-	57,57,58,58	5
2	SO4	A	900	5/5	0.85	0.18	-	58,59,60,61	5

## 6.5 Other polymers [i](#)

There are no such residues in this entry.