

# wwPDB X-ray Structure Validation Summary Report (i)

Apr 12, 2023 – 04:20 pm BST

PDB ID : 8C5P

Title: E. coli NfsB mutant N71S T41L with acetate

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Deposited on : 2023-01-10

Resolution : 1.69 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 : 1.8.4, CSD as541be (2020)

Xtriage (Phenix) : 1.13

EDS : 2.32.2

buster-report : 1.1.7 (2018)

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)

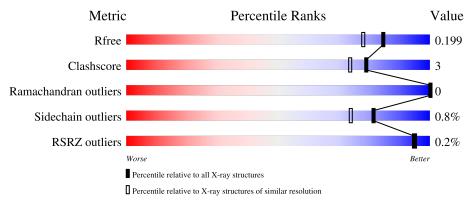
 $\begin{tabular}{lll} Validation Pipeline (wwPDB-VP) & : & 2.32.2 \end{tabular}$ 

## 1 Overall quality at a glance (i)

The following experimental techniques were used to determine the structure:  $X\text{-}RAY\ DIFFRACTION$ 

The reported resolution of this entry is 1.69 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	Similar resolution
Metric	$(\# \mathrm{Entries})$	$(\#  ext{Entries},  ext{ resolution range}( ext{Å}))$
$R_{free}$	130704	4298 (1.70-1.70)
Clashscore	141614	4695 (1.70-1.70)
Ramachandran outliers	138981	4610 (1.70-1.70)
Sidechain outliers	138945	4610 (1.70-1.70)
RSRZ outliers	127900	4222 (1.70-1.70)

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	217	94%	6%
1	В	217	92%	7%

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
ſ	5	ACT	A	607	_	-	X	-



## 2 Entry composition (i)

There are 6 unique types of molecules in this entry. The entry contains 4139 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.

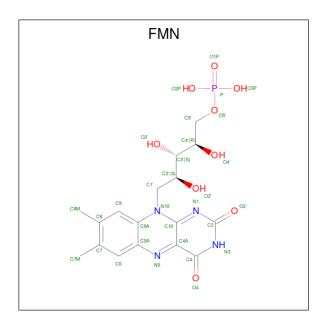
• Molecule 1 is a protein called Oxygen-insensitive NAD(P)H nitroreductase.

Mol	Chain	Residues	Atoms			ZeroOcc	AltConf	Trace		
1	Δ	216	Total	С	N	О	S	0	7	0
1	ı A	210	1736	1102	296	333	5		'	
1	D	216	Total	С	N	О	S	0	0	0
1	Б	210	1749	1110	299	334	6		8   	

There are 4 discrepancies between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	Comment	Reference
A	41	LEU	THR	engineered mutation	UNP P38489
A	71	SER	ASN	engineered mutation	UNP P38489
В	41	LEU	THR	engineered mutation	UNP P38489
В	71	SER	ASN	engineered mutation	UNP P38489

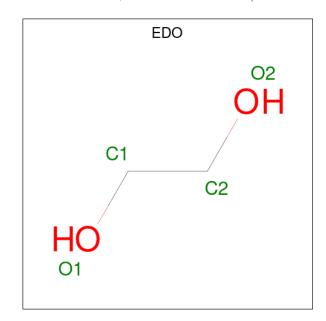
• Molecule 2 is FLAVIN MONONUCLEOTIDE (three-letter code: FMN) (formula:  $C_{17}H_{21}N_4O_9P$ ).





Mol	Chain	Residues	${f Atoms}$			ZeroOcc	AltConf		
2	Λ	1	Total	С	N	О	Р	0	0
	2   A	1	31	17	4	9	1	U	0
2	D	1	Total	С	N	О	Р	0	0
	Б	1	31	17	4	9	1	U	0

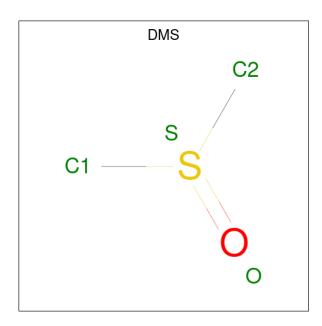
 $\bullet$  Molecule 3 is 1,2-ETHANEDIOL (three-letter code: EDO) (formula:  $\mathrm{C_2H_6O_2}).$ 



Mol	Chain	Residues	Atoms	ZeroOcc	AltConf
3	A	1	Total C O 8 4 4	0	1
3	A	1	Total C O 4 2 2	0	0
3	A	1	Total C O 4 2 2	0	0
3	В	1	Total C O 8 4 4	0	1
3	В	1	Total C O 4 2 2	0	0

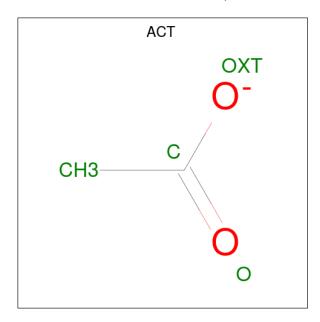
 $\bullet$  Molecule 4 is DIMETHYL SULFOXIDE (three-letter code: DMS) (formula:  $\mathrm{C_2H_6OS}).$ 





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

 $\bullet$  Molecule 5 is ACETATE ION (three-letter code: ACT) (formula:  $\mathrm{C_2H_3O_2}).$ 



Mo	Chain	Residues	Atoms	ZeroOcc	AltConf
5	A	1	Total C O 4 2 2	0	0
5	В	1	Total C O 8 4 4	0	1

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Mol	Chain	Residues	Atoms		ZeroOcc	AltConf	
5	В	1	Total 4	C 2	O 2	0	0

### • Molecule 6 is water.

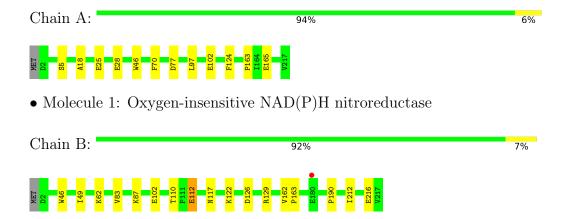
Mol	Chain	Residues	Atoms	ZeroOcc	AltConf
6	A	281	Total O 289 289	0	8
6	В	241	Total O 251 251	0	10



# 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: Oxygen-insensitive NAD(P)H nitroreductase





# 4 Data and refinement statistics (i)

Property	Value	Source
Space group	P 41 21 2	Depositor
Cell constants	57.22Å 57.22Å 262.94Å	Depositor
a, b, c, $\alpha$ , $\beta$ , $\gamma$	90.00° 90.00° 90.00°	Depositor
Resolution (Å)	43.16 - 1.69	Depositor
rtesolution (A)	43.16 - 1.69	EDS
% Data completeness	99.0 (43.16-1.69)	Depositor
(in resolution range)	99.1 (43.16-1.69)	EDS
$R_{merge}$	(Not available)	Depositor
$R_{sym}$	0.05	Depositor
$< I/\sigma(I) > 1$	2.08 (at 1.70Å)	Xtriage
Refinement program	REFMAC 5	Depositor
D D.	0.150 , 0.188	Depositor
$R, R_{free}$	0.163 , 0.199	DCC
$R_{free}$ test set	2499 reflections (5.04%)	wwPDB-VP
Wilson B-factor (Å <sup>2</sup> )	15.1	Xtriage
Anisotropy	0.339	Xtriage
Bulk solvent $k_{sol}(e/Å^3)$ , $B_{sol}(Å^2)$	0.34, 51.0	EDS
L-test for twinning <sup>2</sup>	$ < L > = 0.46, < L^2> = 0.29$	Xtriage
Estimated twinning fraction	No twinning to report.	Xtriage
$F_o, F_c$ correlation	0.96	EDS
Total number of atoms	4139	wwPDB-VP
Average B, all atoms (Å <sup>2</sup> )	19.0	wwPDB-VP

Xtriage's analysis on translational NCS is as follows: The largest off-origin peak in the Patterson function is 3.92% of the height of the origin peak. No significant pseudotranslation is detected.

<sup>&</sup>lt;sup>2</sup>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.



<sup>&</sup>lt;sup>1</sup>Intensities 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: FMN, ACT, DMS, EDO

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	Bor RMSZ	nd lengths	Bond angles		
IVIOI	Mol Chain		# Z  > 5	RMSZ	# Z  > 5	
1	A	0.76	$3/1771 \ (0.2\%)$	0.89	1/2393 (0.0%)	
1	В	0.74	2/1784~(0.1%)	0.90	0/2409	
All	All	0.75	5/3555 (0.1%)	0.89	1/4802 (0.0%)	

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	В	0	1

#### All (5) bond length outliers are listed below:

Mol	Chain	$\operatorname{Res}$	Type	Atoms	$\mathbf{Z}$	$\operatorname{Observed}(\text{\AA})$	$\operatorname{Ideal}( ext{\AA})$
1	В	216	GLU	CD-OE2	-6.84	1.18	1.25
1	В	102	GLU	CD-OE2	6.38	1.32	1.25
1	A	25	GLU	CD-OE2	-6.25	1.18	1.25
1	A	102	GLU	CD-OE2	5.49	1.31	1.25
1	A	165	GLU	CD-OE2	5.18	1.31	1.25

#### All (1) bond angle outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	$Observed(^o)$	$\operatorname{Ideal}({}^{o})$
1	A	25	GLU	OE1-CD-OE2	-5.19	117.07	123.30

There are no chirality outliers.

All (1) planarity outliers are listed below:



$\mathbf{M}$	ol	Chain	Res	Type	Group
1	-	В	129	ARG	Sidechain

## 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	1736	0	1719	6	1
1	В	1749	0	1737	10	0
2	A	31	0	19	1	0
2	В	31	0	19	1	0
3	A	16	0	24	2	0
3	В	12	0	18	2	0
4	A	8	0	12	0	0
5	A	4	0	3	3	0
5	В	12	0	9	1	0
6	A	289	0	0	4	0
6	В	251	0	0	3	0
All	All	4139	0	3560	20	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 3.

The worst 5 of 20 close contacts within the same asymmetric unit are listed below, sorted by their clash magnitude.

Atom-1	Atom-2	Interatomic	Clash
Atom-1	Atom-2	${ m distance}({ m \AA})$	overlap (Å)
5:B:303[B]:ACT:O	6:B:401:HOH:O	2.15	0.63
5:A:607:ACT:H1	6:A:732:HOH:O	2.00	0.59
1:B:117:ASN:HD21	3:B:302[B]:EDO:H12	1.67	0.58
1:A:124:PHE:HB3	5:A:607:ACT:H3	1.91	0.52
3:A:606:EDO:H21	6:A:715:HOH:O	2.11	0.49

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	$\begin{array}{c} {\rm Interatomic} \\ {\rm distance} \ ({\rm \AA}) \end{array}$	Clash overlap (Å)
1:A:5[B]:SER:OG	1:A:28:GLU:OE1[5_545]	2.16	0.04



## 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	Perce	$\mathbf{ntiles}$
1	A	221/217 (102%)	219 (99%)	2 (1%)	0	100	100
1	В	$223/217 \ (103\%)$	221 (99%)	2 (1%)	0	100	100
All	All	444/434 (102%)	440 (99%)	4 (1%)	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	Percentiles		
1	A	185/179 (103%)	184 (100%)	1 (0%)	88 83	
1	В	187/179 (104%)	185 (99%)	2 (1%)	73 63	
All	All	372/358 (104%)	369 (99%)	3 (1%)	81 74	

All (3) residues with a non-rotameric sidechain are listed below:

Mol	Chain	Res	Type
1	A	46	TRP
1	В	46	TRP
1	В	112	GLU

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)

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)

15 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).

N / - 1	Т	Chain	Res	Link	Во	ond leng	ths	В	ond ang	les
Mol	Type	Chain	nes	Lilik	Counts	RMSZ	# Z  > 2	Counts	RMSZ	$\mid \# Z  > 2$
3	EDO	A	602[A]	-	3,3,3	1.03	0	2,2,2	0.38	0
3	EDO	A	606	-	3,3,3	0.19	0	2,2,2	0.50	0
3	EDO	В	302[A]	-	3,3,3	0.32	0	2,2,2	0.44	0
5	ACT	В	305	-	3,3,3	0.64	0	3,3,3	1.31	0
3	EDO	В	304	-	3,3,3	0.54	0	2,2,2	0.69	0
4	DMS	A	603	-	3,3,3	0.52	0	3,3,3	0.27	0
3	EDO	A	602[B]	-	3,3,3	0.68	0	2,2,2	0.28	0
3	EDO	В	302[B]	-	3,3,3	0.25	0	2,2,2	0.44	0
5	ACT	В	303[A]	-	3,3,3	0.48	0	3,3,3	1.54	1 (33%)
2	FMN	A	601	-	33,33,33	1.18	4 (12%)	48,50,50	0.81	2 (4%)
3	EDO	A	604	-	3,3,3	0.19	0	2,2,2	0.54	0
5	ACT	A	607	-	3,3,3	0.71	0	3,3,3	0.92	0
4	DMS	A	605	-	3,3,3	0.68	0	3,3,3	0.19	0
5	ACT	В	303[B]	-	3,3,3	0.58	0	3,3,3	1.41	0
2	FMN	В	301	-	33,33,33	1.19	5 (15%)	48,50,50	0.89	1 (2%)

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	EDO	A	602[A]	-	-	1/1/1/1	-
3	EDO	A	606	-	-	1/1/1/1	-
3	EDO	В	302[A]	-	-	0/1/1/1	-
3	EDO	В	304	_	-	1/1/1/1	-
3	EDO	A	602[B]	-	-	0/1/1/1	-
3	EDO	В	302[B]	-	-	1/1/1/1	-
2	FMN	A	601	-	-	2/18/18/18	0/3/3/3
3	EDO	A	604	-	-	1/1/1/1	-
2	FMN	В	301	-	-	2/18/18/18	0/3/3/3

The worst 5 of 9 bond length outliers are listed below:

Mol	Chain	Res	Type	Atoms	$\mathbf{Z}$	$\operatorname{Observed}(\text{\AA})$	Ideal(Å)
2	A	601	FMN	O2-C2	2.98	1.29	1.24
2	В	301	FMN	O3'-C3'	2.66	1.49	1.43
2	В	301	FMN	C2-N3	-2.54	1.33	1.39
2	В	301	FMN	C9-C8	-2.49	1.35	1.39
2	A	601	FMN	C1'-C2'	-2.40	1.49	1.52

All (4) bond angle outliers are listed below:

Mol	Chain	Res	Type	Atoms	$\mathbf{Z}$	$\mathbf{Observed}(^o)$	$\operatorname{Ideal}({}^{o})$
2	A	601	FMN	O4'-C4'-C3'	2.35	114.81	109.10
5	В	303[A]	ACT	O-C-CH3	-2.14	114.00	122.33
2	A	601	FMN	C5A-N5-C4A	-2.13	114.54	118.07
2	В	301	FMN	C5A-N5-C4A	-2.07	114.63	118.07

There are no chirality outliers.

5 of 9 torsion outliers are listed below:

Mol	Chain	Res	Type	Atoms
2	A	601	FMN	C2'-C1'-N10-C10
2	В	301	FMN	C2'-C1'-N10-C10
3	A	604	EDO	O1-C1-C2-O2
3	В	302[B]	EDO	O1-C1-C2-O2
3	A	606	EDO	O1-C1-C2-O2

There are no ring outliers.

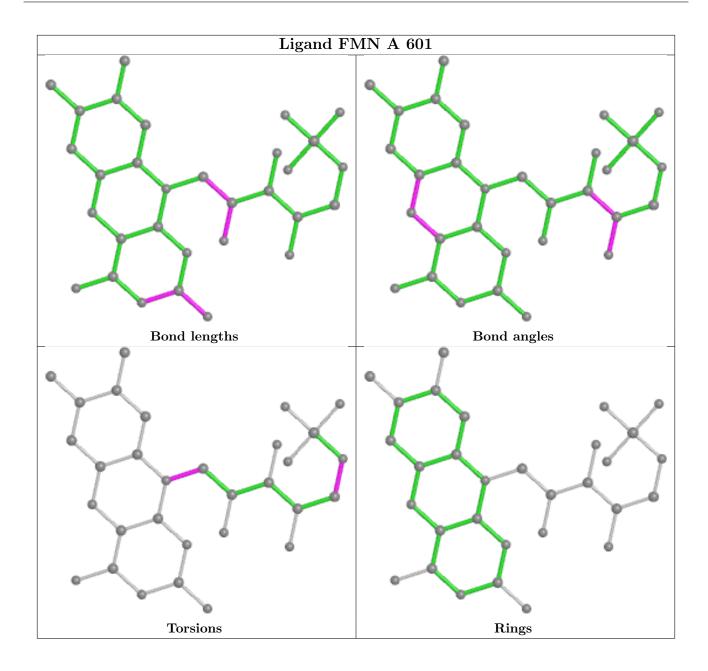


6 monomers are involved in 10 short contacts:

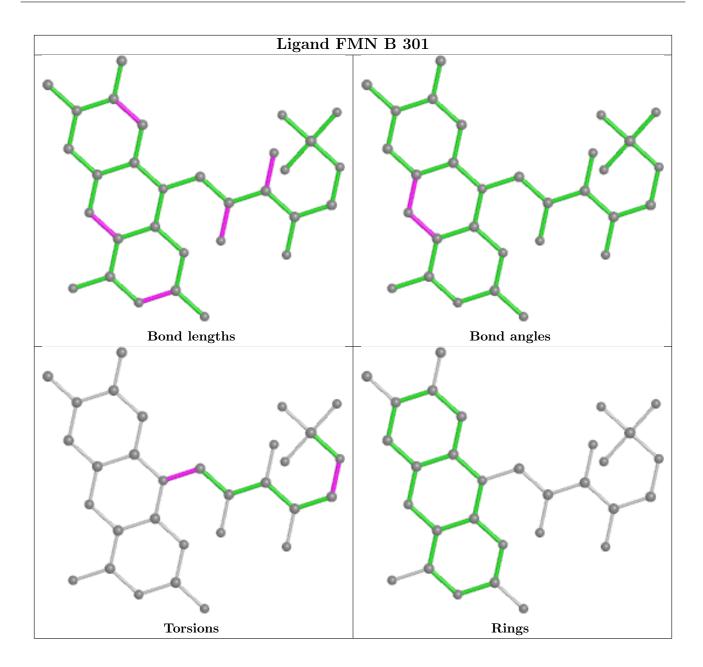
Mol	Chain	Res	Type	Clashes	Symm-Clashes
3	A	606	EDO	2	0
3	В	302[B]	EDO	2	0
2	A	601	FMN	1	0
5	A	607	ACT	3	0
5	В	303[B]	ACT	1	0
2	В	301	FMN	1	0

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 > 2	$OWAB(A^2)$	Q<0.9
1	A	216/217 (99%)	-0.38	0 100 100	7, 14, 29, 41	0
1	В	216/217 (99%)	-0.24	1 (0%) 91 92	7, 18, 32, 50	0
All	All	432/434 (99%)	-0.31	1 (0%) 95 95	7, 16, 31, 50	0

#### All (1) RSRZ outliers are listed below:

Mol	Chain	Res	Type	RSRZ
1	В	180	GLU	2.7

## 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	$\mathbf{B} ext{-}\mathbf{factors}(\mathbf{\mathring{A}}^2)$	Q<0.9
4	DMS	A	605	4/4	0.79	0.23	46,51,52,60	0
3	EDO	В	304	4/4	0.80	0.14	47,49,49,50	0
3	EDO	A	604	4/4	0.88	0.17	42,42,45,45	0
5	ACT	A	607	4/4	0.89	0.13	34,35,38,47	3

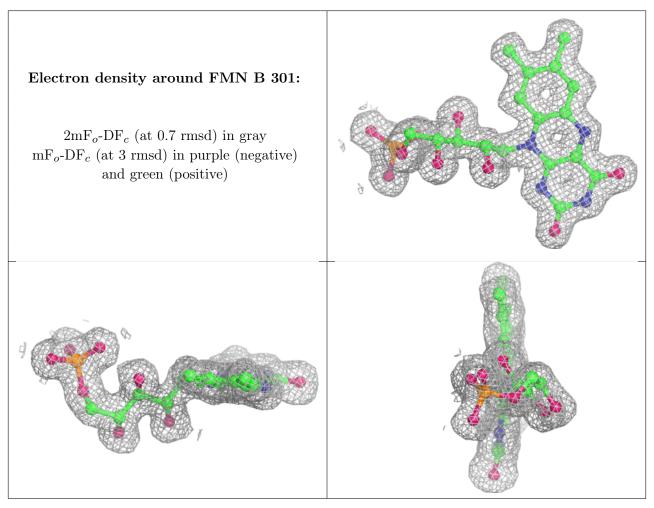
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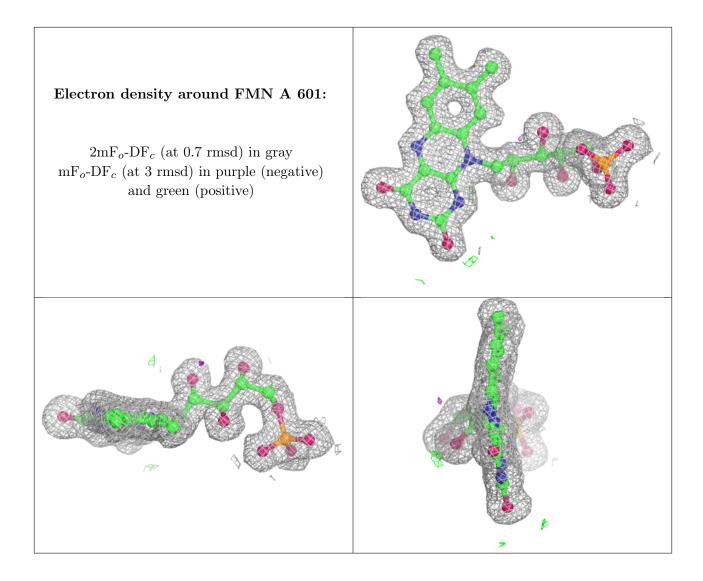
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Mol	Type	Chain	Res	Atoms	RSCC	RSR	$ m B ext{-}factors(\AA^2)$	Q < 0.9
5	ACT	В	303[A]	4/4	0.90	0.33	9,11,12,12	4
5	ACT	В	303[B]	4/4	0.90	0.33	33,33,36,37	4
3	EDO	В	302[B]	4/4	0.91	0.15	22,26,26,30	4
3	EDO	A	606	4/4	0.91	0.11	36,42,46,47	0
3	EDO	В	302[A]	4/4	0.91	0.15	18,20,22,23	4
4	DMS	A	603	4/4	0.92	0.15	42,43,48,49	0
3	EDO	A	602[A]	4/4	0.94	0.23	31,35,38,39	4
3	EDO	A	602[B]	4/4	0.94	0.23	18,25,32,42	4
5	ACT	В	305	4/4	0.94	0.15	23,25,30,36	3
2	FMN	В	301	31/31	0.98	0.06	9,12,17,22	0
2	FMN	A	601	31/31	0.98	0.07	7,8,11,15	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.







## 6.5 Other polymers (i)

There are no such residues in this entry.

