

wwPDB X-ray Structure Validation Summary Report (i)

Feb 29, 2024 – 08:26 AM EST

PDB ID	:	5TZO
Title	:	Computationally Designed Fentanyl Binder - Fen49*-Complex
Authors	:	Bick, M.J.; Greisen, P.J.; Morey, K.J.; Antunes, M.S.; La, D.; Sankaran, B.;
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Deposited on	:	2016-11-22
Resolution	:	1.67 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:

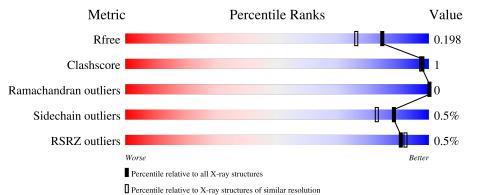
MolProbity Mogul Xtriage (Phenix) EDS	:	4.02b-467 1.8.5 (274361), CSD as541be (2020) 1.13 2.36
buster-report Percentile statistics Refmac	: : :	1.1.7 (2018) 20191225.v01 (using entries in the PDB archive December 25th 2019) 5.8.0158 7.0.044 (Gargrove)
Ideal geometry (DNA, RNA) Validation Pipeline (wwPDB-VP)		Parkinson et al. (1996) 2.36

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 1.67 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	$\begin{array}{c} \textbf{Whole archive} \\ (\#\textbf{Entries}) \end{array}$	${f Similar\ resolution}\ (\#{ m Entries,\ resolution\ range}({ m \AA}))$
R_{free}	130704	6780 (1.70-1.66)
Clashscore	141614	7310 (1.70-1.66)
Ramachandran outliers	138981	7173 (1.70-1.66)
Sidechain outliers	138945	7172 (1.70-1.66)
RSRZ outliers	127900	6661 (1.70-1.66)

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	А	188	% 95% • •	•
1	В	188	96%	_
1	С	188	% • • 98% •	•



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2 Entry composition (i)

There are 5 unique types of molecules in this entry. The entry contains 9485 atoms, of which 4205 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.

Mol	Chain	Residues		Atoms					ZeroOcc	AltConf	Trace
1	1 Λ	185	Total	С	Η	Ν	0	\mathbf{S}	0	12	0
1	Л		2827	930	1346	253	296	2	0		
1	В	185	Total	С	Η	Ν	Ο	\mathbf{S}	0	11	0
	D	100	2846	931	1363	256	293	3	0	11	0
1	С	185	Total	С	Η	Ν	0	S	0	8	0
I U	100	2799	920	1340	252	285	2	0	0	0	

• Molecule 1 is a protein called Endo-1,4-beta-xylanase A.

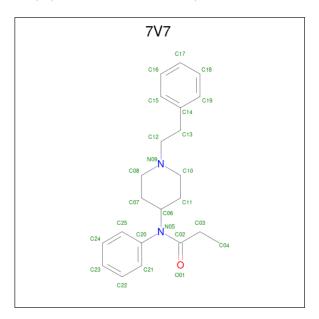
Chain	Residue	Modelled	Actual	Comment	Reference
А	-2	GLY	-	expression tag	UNP P18429
А	-1	PRO	-	expression tag	UNP P18429
А	0	HIS	-	expression tag	UNP P18429
А	1	MET	-	expression tag	UNP P18429
А	7	LEU	GLN	conflict	UNP P18429
А	9	PHE	TRP	conflict	UNP P18429
А	35	SER	ASN	conflict	UNP P18429
А	63	TRP	ASN	conflict	UNP P18429
А	65	ALA	TYR	conflict	UNP P18429
А	67	ALA	THR	conflict	UNP P18429
А	69	VAL	TYR	conflict	UNP P18429
А	78	ALA	GLU	conflict	UNP P18429
А	88	ALA	TYR	conflict	UNP P18429
А	90	TRP	PRO	conflict	UNP P18429
А	172	ALA	GLU	conflict	UNP P18429
В	-2	GLY	-	expression tag	UNP P18429
В	-1	PRO	-	expression tag	UNP P18429
В	0	HIS	-	expression tag	UNP P18429
В	1	MET	-	expression tag	UNP P18429
В	7	LEU	GLN	conflict	UNP P18429
В	9	PHE	TRP	conflict	UNP P18429
В	35	SER	ASN	conflict	UNP P18429
В	63	TRP	ASN	conflict	UNP P18429

There are 45 discrepancies between the modelled and reference sequences:



Chain	Residue	Modelled	Actual	Comment	Reference
В	65	ALA	TYR	conflict	UNP P18429
В	67	ALA	THR	conflict	UNP P18429
В	69	VAL	TYR	conflict	UNP P18429
В	78	ALA	GLU	conflict	UNP P18429
В	88	ALA	TYR	conflict	UNP P18429
В	90	TRP	PRO	conflict	UNP P18429
В	172	ALA	GLU	conflict	UNP P18429
С	-2	GLY	-	expression tag	UNP P18429
С	-1	PRO	-	expression tag	UNP P18429
С	0	HIS	-	expression tag	UNP P18429
С	1	MET	-	expression tag	UNP P18429
С	7	LEU	GLN	conflict	UNP P18429
С	9	PHE	TRP	conflict	UNP P18429
С	35	SER	ASN	conflict	UNP P18429
С	63	TRP	ASN	conflict	UNP P18429
С	65	ALA	TYR	conflict	UNP P18429
С	67	ALA	THR	conflict	UNP P18429
С	69	VAL	TYR	conflict	UNP P18429
С	78	ALA	GLU	conflict	UNP P18429
С	88	ALA	TYR	conflict	UNP P18429
С	90	TRP	PRO	conflict	UNP P18429
С	172	ALA	GLU	conflict	UNP P18429

• Molecule 2 is N-phenyl-N-[1-(2-phenylethyl)piperidin-4-yl]propanamide (three-letter code: 7V7) (formula: $C_{22}H_{28}N_2O$).





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Mol	Chain	Residues		Ate	oms			ZeroOcc	AltConf	
2	А	1	Total	С	Η	Ν	0	0	0	
	A	1	53	22	28	2	1	0	0	
2	А	1	Total	С	Η	Ν	0	0	0	
	Л	1	53	22	28	2	1	0	0	
2	В	1	Total	С	Η	Ν	Ο	0	0	
	D	1	53	22	28	2	1	0	0	
2	В	1	Total	С	Η	Ν	Ο	0	0	
	D	1	39	15	21	2	1	0	0	
2	С	1	Total	С	Η	Ν	Ο	0	0	
2	U	1	53	22	28	2	1	0	U	
2	С	1	Total	С	Η	Ν	Ο	0	0	
		1	42	16	23	2	1	0	0	

• Molecule 3 is POTASSIUM ION (three-letter code: K) (formula: K).

Mol	Chain	Residues	Atoms	ZeroOcc	AltConf
3	А	2	Total K 2 2	0	0
3	В	1	Total K 1 1	0	0

• Molecule 4 is CHLORIDE ION (three-letter code: CL) (formula: Cl).

Mol	Chain	Residues	Atoms	ZeroOcc	AltConf
4	А	1	Total Cl 1 1	0	0
4	В	1	Total Cl 1 1	0	0
4	С	1	Total Cl 1 1	0	0

• Molecule 5 is water.

Mol	Chain	Residues	Atoms	ZeroOcc	AltConf
5	А	217	Total O 217 217	0	0
5	В	231	Total O 231 231	0	0
5	С	266	Total O 266 266	0	0



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: Endo-1,4-beta-xylanase A

Chain A:	95% •••
GLY PR0 HIS M15 A15 A172 A172 N131 W181	
• Molecule 1: Endo-1,4-beta-xylanase	Α
Chain B:	96%
GLY HIS HIS 133 780 175 4175 4175	
• Molecule 1: Endo-1,4-beta-xylanase	А
Chain C:	98% •
CLV PRO HI S HI B MI PRO HI B S	



4 Data and refinement statistics (i)

Property	Value	Source
Space group	P 21 21 21	Depositor
Cell constants	56.07Å 73.25Å 136.64Å	Depositor
a, b, c, α , β , γ	90.00° 90.00° 90.00°	Depositor
Resolution (Å)	51.87 - 1.67	Depositor
Resolution (A)	51.87 - 1.67	EDS
% Data completeness	98.6 (51.87-1.67)	Depositor
(in resolution range)	98.7(51.87-1.67)	EDS
R _{merge}	0.08	Depositor
R_{sym}	(Not available)	Depositor
$< I/\sigma(I) > 1$	$2.40 (at 1.67 \text{\AA})$	Xtriage
Refinement program	PHENIX dev_2463	Depositor
D D.	0.166 , 0.203	Depositor
R, R_{free}	0.164 , 0.198	DCC
R_{free} test set	3326 reflections $(5.10%)$	wwPDB-VP
Wilson B-factor $(Å^2)$	13.6	Xtriage
Anisotropy	0.226	Xtriage
Bulk solvent $k_{sol}(e/Å^3), B_{sol}(Å^2)$	0.41 , 49.0	EDS
L-test for twinning ²	$ \langle L \rangle = 0.47, \langle L^2 \rangle = 0.30$	Xtriage
Estimated twinning fraction	No twinning to report.	Xtriage
F_o, F_c correlation	0.96	EDS
Total number of atoms	9485	wwPDB-VP
Average B, all atoms $(Å^2)$	17.0	wwPDB-VP

Xtriage's analysis on translational NCS is as follows: The largest off-origin peak in the Patterson function is 6.02% of the height of the origin peak. No significant pseudotranslation is detected.

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.



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: CL, K, $7\mathrm{V}7$

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		
	Unam	RMSZ	# Z > 5	RMSZ	# Z > 5	
1	А	0.36	0/1542	0.64	2/2117~(0.1%)	
1	В	0.37	0/1561	0.60	0/2138	
1	С	0.39	0/1522	0.60	0/2085	
All	All	0.37	0/4625	0.62	2/6340~(0.0%)	

There are no bond length outliers.

All (2) bond angle outliers are listed below:

Mol	Chain	Res	Type	Atoms	Ζ	$Observed(^{o})$	$Ideal(^{o})$
1	А	37	VAL	CG1-CB-CG2	8.03	123.74	110.90
1	А	37	VAL	CA-CB-CG1	5.86	119.68	110.90

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	А	1481	1346	1320	3	0
1	В	1483	1363	1335	3	0
1	С	1459	1340	1316	0	0
2	А	50	56	0	0	0
2	В	43	49	0	0	0
2	С	44	51	0	0	0



Mol	Chain	Non-H	${ m H(model)}$	H(added)	Clashes	Symm-Clashes				
3	А	2	0	0	0	0				
3	В	1	0	0	0	0				
4	А	1	0	0	0	0				
4	В	1	0	0	0	0				
4	С	1	0	0	0	0				
5	А	217	0	0	1	2				
5	В	231	0	0	2	1				
5	С	266	0	0	0	1				
All	All	5280	4205	3971	6	2				

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 1.

The worst 5 of 6 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:A:156:HIS:O	5:A:301:HOH:O	2.15	0.63
1:B:122:ARG:NH2	5:B:302:HOH:O	2.32	0.62
1:B:80:TYR:OH	5:B:301:HOH:O	2.18	0.52
1:A:78:ALA:O	1:A:127:GLN:HA	2.15	0.47
1:A:37:VAL:HG23	1:A:172:ALA:HB2	2.03	0.41

All (2) 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)
5:A:427:HOH:O	5:C:328:HOH:O[2_454]	2.13	0.07
5:A:307:HOH:O	5:B:314:HOH:O[4_545]	2.17	0.03

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	ntiles
1	А	196/188~(104%)	192~(98%)	4(2%)	0	100	100
1	В	196/188~(104%)	191~(97%)	5(3%)	0	100	100
1	С	191/188~(102%)	188 (98%)	3~(2%)	0	100	100
All	All	583/564~(103%)	571 (98%)	12 (2%)	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 Outlier		Percentiles
1	А	155/149~(104%)	153~(99%)	2(1%)	69 54
1	В	158/149~(106%)	158 (100%)	0	100 100
1	С	152/149~(102%)	152 (100%)	0	100 100
All	All	465/447~(104%)	463 (100%)	2 (0%)	88 86

All (2) residues with a non-rotameric sidechain are listed below:

Mol	Chain	Res	Type
1	А	37	VAL
1	А	181	ASN

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)

Of 12 ligands modelled in this entry, 6 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	Iol Type Chain Res		Link	Bo	Bond lengths		Bond angles		les	
MIOI	Mol Type Chain	nes	LIIIK	Counts	RMSZ	# Z >2	Counts	RMSZ	# Z >2	
2	7V7	А	202	-	$27,\!27,\!27$	0.79	1 (3%)	34,35,35	1.08	4 (11%)
2	7V7	С	201	-	$27,\!27,\!27$	0.56	0	34,35,35	0.94	1 (2%)
2	7V7	В	202	-	$19,\!19,\!27$	1.04	2 (10%)	24,25,35	1.20	2 (8%)
2	7V7	В	201	-	$27,\!27,\!27$	0.96	1 (3%)	34,35,35	1.05	3 (8%)
2	7V7	А	201	-	27,27,27	0.80	1 (3%)	34,35,35	1.13	3 (8%)
2	7V7	С	202	-	$20,\!20,\!27$	0.73	0	$25,\!26,\!35$	0.96	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	7V7	А	202	-	-	0/19/29/29	0/3/3/3
2	7V7	С	201	-	-	0/19/29/29	0/3/3/3
2	7V7	В	202	-	-	0/14/24/29	0/2/2/3
2	7V7	В	201	-	-	0/19/29/29	0/3/3/3
2	7V7	А	201	-	-	0/19/29/29	0/3/3/3
2	7V7	С	202	-	-	2/16/26/29	0/2/2/3

All (5) bond length outliers are listed below:

Mol	Chain	Res	Type	Atoms	Ζ	Observed(Å)	Ideal(Å)
2	В	201	7V7	C02-N05	2.95	1.41	1.37
2	В	202	7V7	C11-C06	2.43	1.58	1.52



	Chain	-	10	Atoms	Ζ	Observed(Å)	Ideal(Å)
2	А	202	7V7	C02-N05	2.23	1.40	1.37
2	А	201	7V7	C02-N05	2.14	1.40	1.37
2	В	202	7V7	C02-N05	2.01	1.40	1.37

The worst 5 of 13 bond angle outliers are listed below:

Mol	Chain	Res	Type	ype Atoms		$Observed(^{o})$	$Ideal(^{o})$
2	В	202	7V7	C03-C02-N05	2.77	122.80	118.00
2	В	202	7V7	O01-C02-C03	-2.50	116.10	121.42
2	В	201	7V7	C11-C10-N09	2.47	114.94	111.11
2	А	202	7V7	C11-C10-N09	2.40	114.84	111.11
2	А	202	7V7	C08-N09-C10	2.40	114.23	108.83

There are no chirality outliers.

All (2) torsion outliers are listed below:

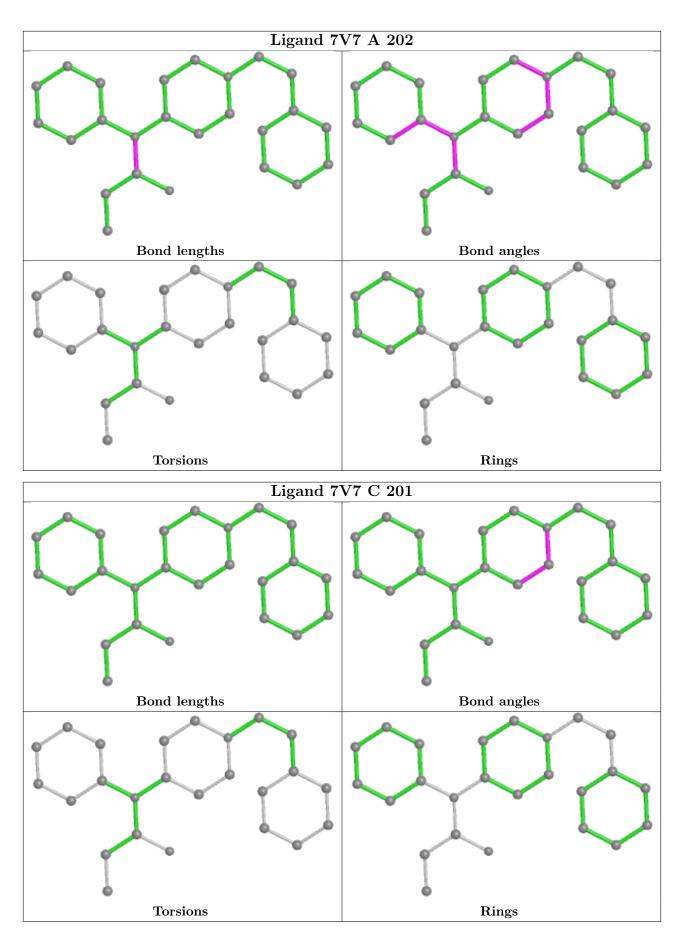
Mol	Chain	Res	Type	Atoms
2	С	202	7V7	C13-C12-N09-C10
2	С	202	7V7	C13-C12-N09-C08

There are no ring outliers.

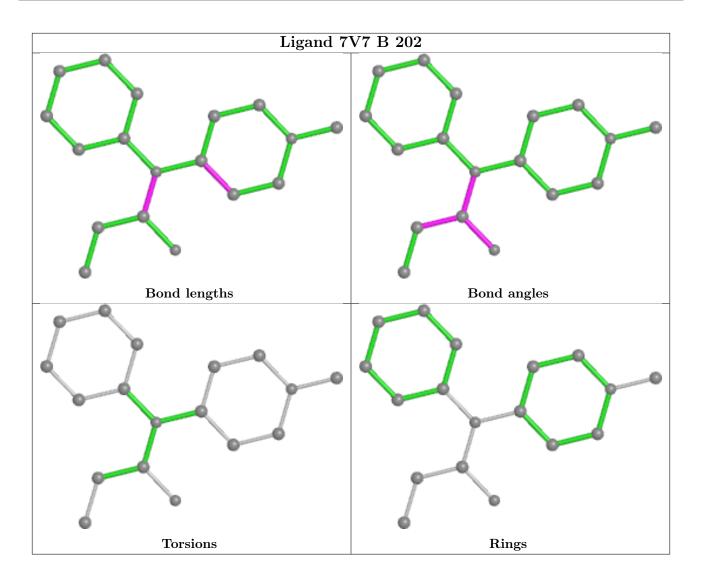
No monomer is involved in short contacts.

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

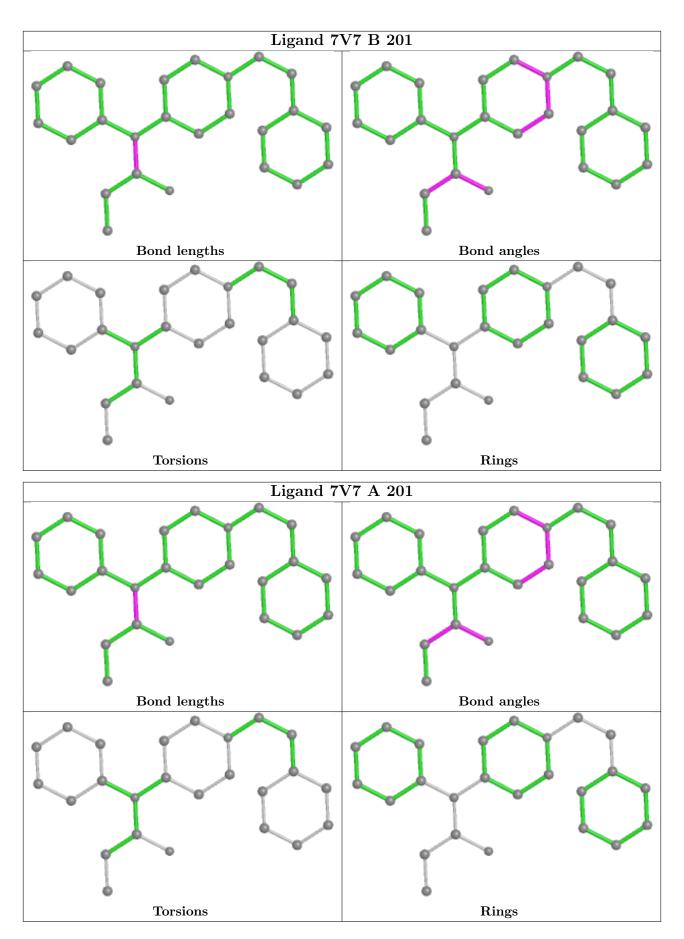




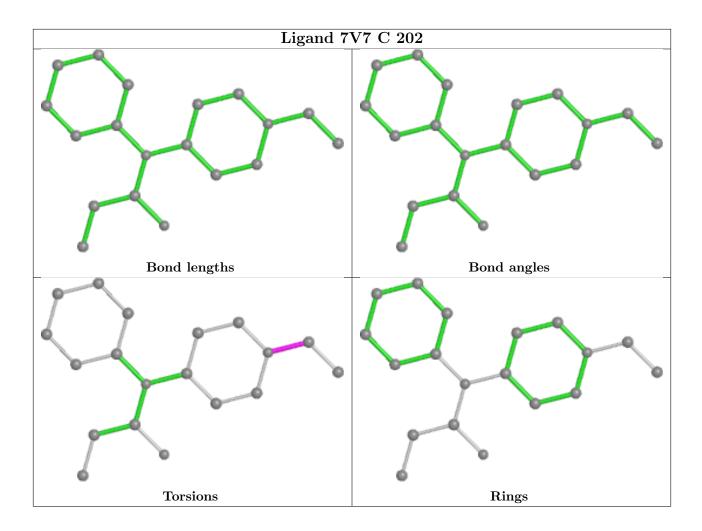












5.7 Other polymers (i)

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	$\langle RSRZ \rangle$	#RSRZ>2	$OWAB(Å^2)$	Q<0.9
1	А	185/188~(98%)	-0.37	2 (1%) 80 83	8, 14, 27, 45	0
1	В	185/188~(98%)	-0.43	0 100 100	6, 12, 28, 41	0
1	С	185/188~(98%)	-0.46	1 (0%) 91 92	6, 11, 24, 44	0
All	All	555/564~(98%)	-0.42	3 (0%) 91 92	6, 12, 27, 45	0

All (3) RSRZ outliers are listed below:

Mol	Chain	Res	Type	RSRZ
1	А	1	MET	3.7
1	С	1	MET	3.2
1	А	113	TYR	2.3

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)

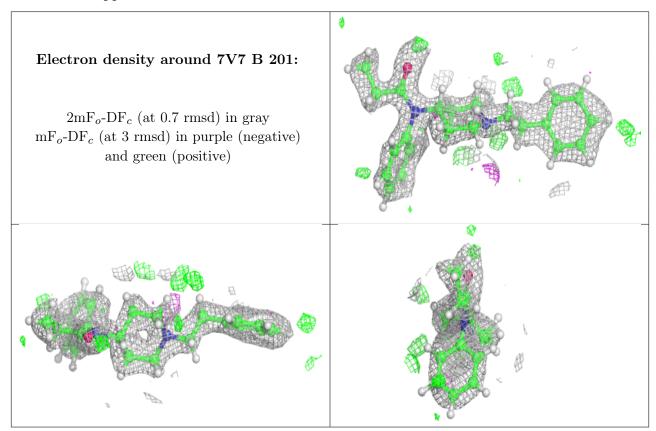
In the following table, the Atoms column lists the number of modelled atoms in the group and the number defined in the chemical component dictionary. The B-factors column lists the minimum, median, 95^{th} percentile and maximum values of B factors of atoms in the group. The column labelled 'Q< 0.9'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

Mol	Type	Chain	Res	Atoms	RSCC	RSR	$\mathbf{B} ext{-factors}(\mathrm{\AA}^2)$	Q < 0.9
2	7V7	В	201	25/25	0.82	0.16	17,26,39,41	0

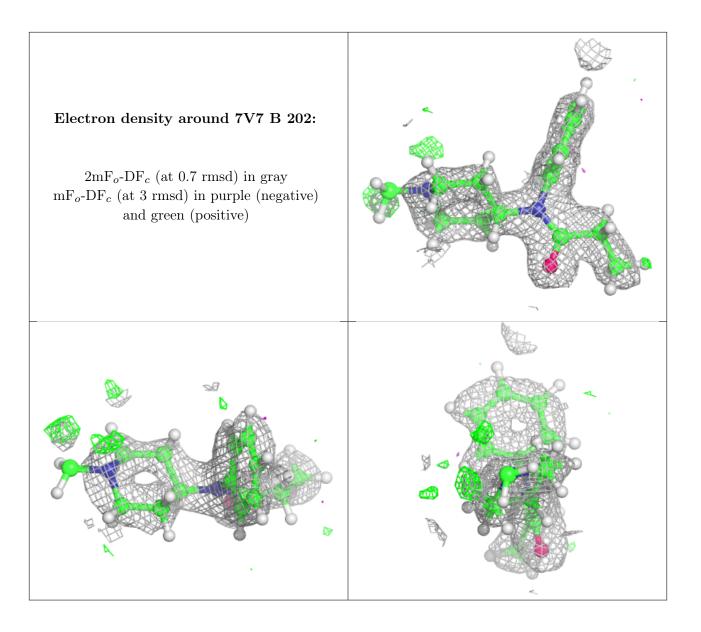


Mol	Type	Chain	Res	Atoms	RSCC	RSR	B -factors($Å^2$)	Q<0.9
2	7V7	В	202	18/25	0.88	0.14	$19,\!29,\!51,\!61$	0
2	7V7	С	202	19/25	0.92	0.10	12,19,40,51	0
2	7V7	С	201	25/25	0.93	0.09	11,17,25,30	0
2	7V7	А	201	25/25	0.94	0.11	$9,\!18,\!55,\!58$	0
2	7V7	А	202	25/25	0.94	0.09	11,19,23,31	0
4	CL	В	203	1/1	0.98	0.08	22,22,22,22	0
3	Κ	А	204	1/1	0.99	0.04	18,18,18,18	0
3	Κ	А	203	1/1	0.99	0.05	16, 16, 16, 16	0
4	CL	А	205	1/1	1.00	0.03	14,14,14,14	0
3	К	В	204	1/1	1.00	0.04	$15,\!15,\!15,\!15$	0
4	CL	С	203	1/1	1.00	0.04	14,14,14,14	0

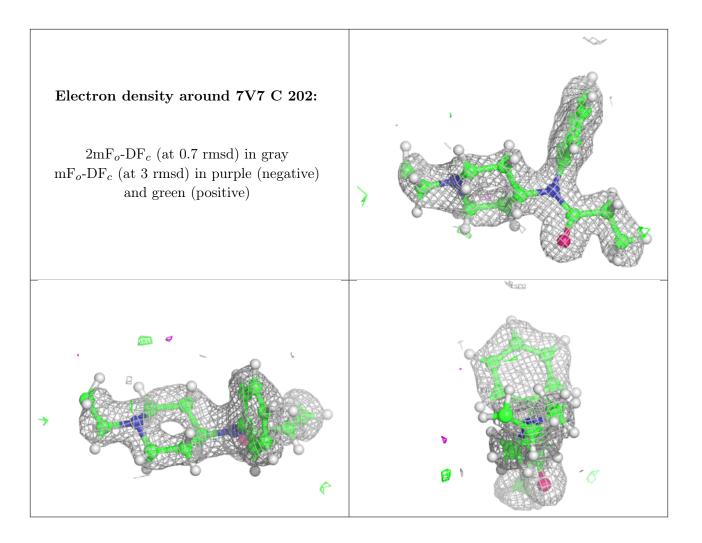
The following is a graphical depiction of the model fit to experimental electron density of all instances of the Ligand of Interest. In addition, ligands with molecular weight > 250 and outliers as shown on the geometry validation Tables will also be included. Each fit is shown from different orientation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



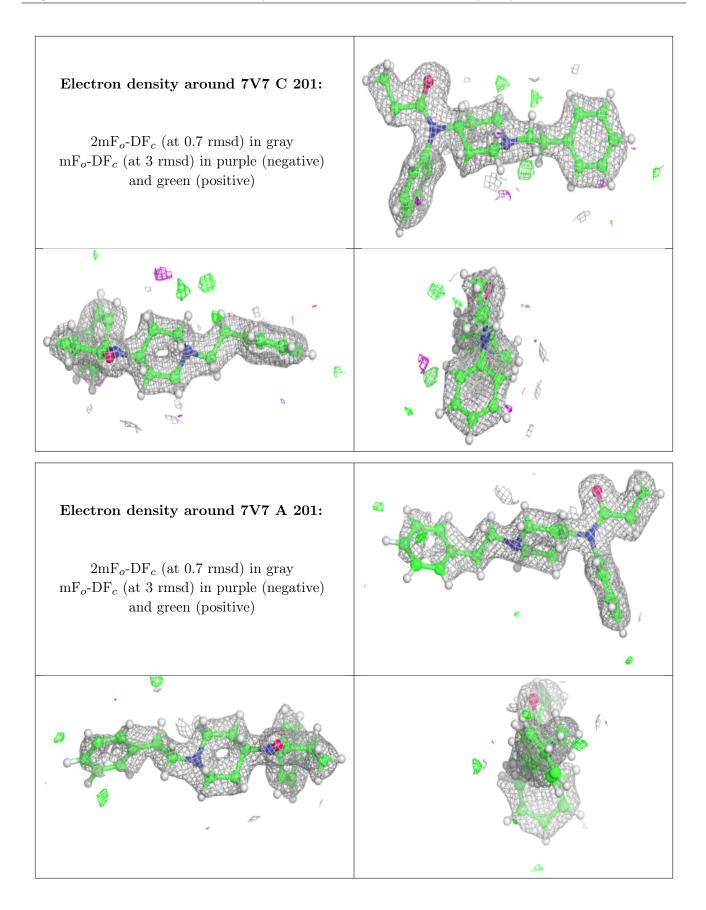




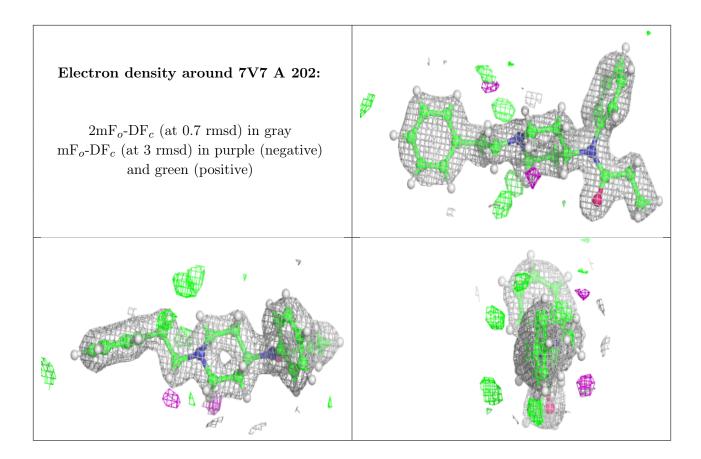












6.5 Other polymers (i)

There are no such residues in this entry.

