

wwPDB X-ray Structure Validation Summary Report (i)

May 1, 2023 – 12:13 PM EDT

PDB ID 8GDS : CRYSTAL STRUCTURE OF RHO-ASSOCIATED PROTEIN KINASE 2 Title : (ROCK2) IN COMPLEX WITH 4-(4-(2-(2,3-DIHYDRO-1H- INDOL-1-YL)-2-OXOETHYL)PHENYL)-1(2H)-PHTHALAZINONE Authors Muckelbauer, J.K. • Deposited on 2023-03-06 2.71 Å(reported) Resolution :

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 (i)) were used in the production of this report:

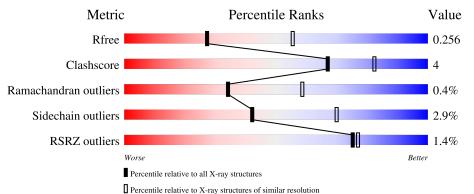
MolProbity		4 09b 467
÷		
Mogul	:	1.8.5 (274361), CSD as541be (2020)
Xtriage (Phenix)	:	1.13
EDS	:	2.32.2
buster-report	:	1.1.7 (2018)
Percentile statistics	:	20191225.v01 (using entries in the PDB archive December 25th 2019)
Refmac	:	5.8.0158
CCP4	:	7.0.044 (Gargrove)
Ideal geometry (proteins)	:	Engh & Huber (2001)
Ideal geometry (DNA, RNA)	:	Parkinson et al. (1996)
Validation Pipeline (wwPDB-VP)	:	2.32.2

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 2.71 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	3359(2.74-2.70)
Clashscore	141614	$3686\ (2.74-2.70)$
Ramachandran outliers	138981	3622(2.74-2.70)
Sidechain outliers	138945	3623 (2.74-2.70)
RSRZ outliers	127900	3276 (2.74-2.70)

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	А	396	88%	9%	•••			
1	В	396	2% 85%	11%				
1	С	396	2% 86%	10%				
1	D	396	2% 84%	12%	·			



2 Entry composition (i)

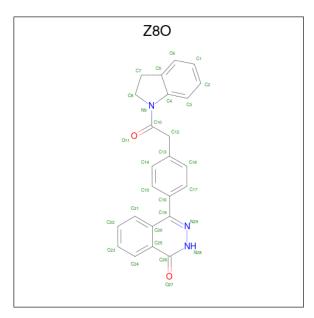
There are 4 unique types of molecules in this entry. The entry contains 12153 atoms, of which 0 are hydrogens and 0 are deuteriums.

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.

Mol	Chain	Residues	Atoms					ZeroOcc	AltConf	Trace
1	А	384	Total	С	Ν	0	\mathbf{S}	0	0	0
	A	304	2919	1881	477	543	18	0	0	0
1	В	384	Total	С	Ν	0	S	0	0	0
	D	304	3004	1930	499	556	19	0		
1	С	385	Total	С	Ν	0	S	0	0	0
	U	000	3008	1938	497	554	19	0	0	
1	Л	379	Total	С	Ν	0	S	0	0	0
	D	519	2978	1923	490	546	19	0		0

• Molecule 1 is a protein called Rho-associated protein kinase 2.

• Molecule 2 is $4-\{4-[2-(2,3-dihydro-1H-indol-1-yl)-2-oxoethyl]phenyl\}phthalazin-1(2H)-one (three-letter code: Z8O) (formula: C₂₄H₁₉N₃O₂) (labeled as "Ligand of Interest" by depositor).$

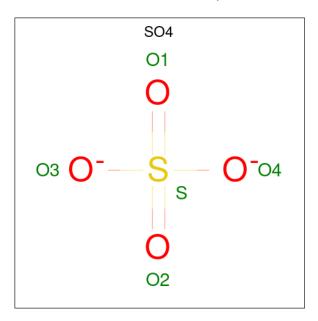


Mol	Chain	Residues	A	Aton	ns		ZeroOcc	AltConf
2	А	1	Total 29	C 24		O 2	0	0



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Mol	Chain	Residues	Atoms	ZeroOcc	AltConf							
2	В	1	Total C N O 29 24 3 2	0	0							
2	С	1	Total C N O 29 24 3 2	0	0							
2	D	1	Total C N O 29 24 3 2	0	0							

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Mol	Chain	Residues	Atom	s	ZeroOcc	AltConf
3	А	1	Total O	S	0	0
0	11	1	5 4	1	0	0
3	А	1	Total O		0	0
		1	5 4	1	Ŭ	
3	А	1	Total O		0	0
		-	5 4	1	Ŭ	0
3	А	1	Total O		0	0
		-	5 4	1	Ŭ	Ŭ
3	А	1	Total O		0	0
		-	5 4	1		<u> </u>
3	В	1	Total O		0	0
		-	5 4	1	Ŭ	0
3	В	1	Total O		0	0
		*	5 4	1	Ŭ	, in the second
3	3 B	B 1	Total O	~	0	0
	5	Ĩ	5 4	1		



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Mol	Chain	Residues	Atoms	ZeroOcc	AltConf
3	С	1	$\begin{array}{ccc} \text{Total} & \text{O} & \text{S} \\ 5 & 4 & 1 \end{array}$	0	0
3	С	1	$\begin{array}{ccc} \text{Total} & \text{O} & \text{S} \\ 5 & 4 & 1 \end{array}$	0	0
3	С	1	$\begin{array}{ccc} \text{Total} & \text{O} & \text{S} \\ 5 & 4 & 1 \end{array}$	0	0
3	С	1	$\begin{array}{ccc} \text{Total} & \text{O} & \text{S} \\ 5 & 4 & 1 \end{array}$	0	0
3	С	1	$\begin{array}{ccc} \text{Total} & \text{O} & \text{S} \\ 5 & 4 & 1 \end{array}$	0	0
3	С	1	$\begin{array}{ccc} \text{Total} & \text{O} & \text{S} \\ 5 & 4 & 1 \end{array}$	0	0
3	D	1	$\begin{array}{ccc} \text{Total} & \text{O} & \text{S} \\ 5 & 4 & 1 \end{array}$	0	0
3	D	1	$\begin{array}{ccc} \text{Total} & \text{O} & \text{S} \\ 5 & 4 & 1 \end{array}$	0	0
3	D	1	$\begin{array}{ccc} \text{Total} & \text{O} & \text{S} \\ 5 & 4 & 1 \end{array}$	0	0

• Molecule 4 is water.

Mol	Chain	Residues	Atoms	ZeroOcc	AltConf
4	А	12	Total O 12 12	0	0
4	В	12	Total O 12 12	0	0
4	С	10	Total O 10 10	0	0
4	D	9	Total O 9 9	0	0



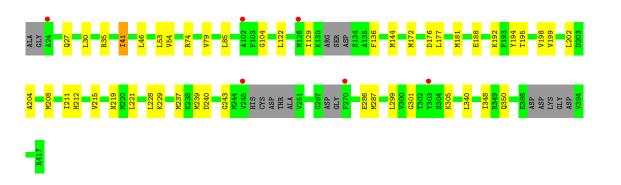
3 Residue-property plots (i)

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: Rho-associated protein kinase 2







4 Data and refinement statistics (i)

Property	Value	Source
Space group	P 21 21 2	Depositor
Cell constants	143.08Å 143.33Å 134.36Å	Depositor
a, b, c, α , β , γ	90.00° 90.00° 90.00°	Depositor
Resolution (Å)	98.03 - 2.71	Depositor
Resolution (A)	101.26 - 2.71	EDS
% Data completeness	69.1 (98.03-2.71)	Depositor
(in resolution range)	$69.1 \ (101.26-2.71)$	EDS
R _{merge}	(Not available)	Depositor
R _{sym}	(Not available)	Depositor
$< I/\sigma(I) > 1$	$1.47 (at 2.69 \text{\AA})$	Xtriage
Refinement program	BUSTER	Depositor
R, R_{free}	0.217 , 0.248	Depositor
Λ, Λ_{free}	0.233 , 0.256	DCC
R_{free} test set	566 reflections (1.08%)	wwPDB-VP
Wilson B-factor $(Å^2)$	53.2	Xtriage
Anisotropy	0.055	Xtriage
Bulk solvent $k_{sol}(e/Å^3)$, $B_{sol}(Å^2)$	0.34, 58.3	EDS
L-test for twinning ²	$< L >=0.48, < L^2>=0.30$	Xtriage
Estimated twinning fraction	0.017 for k,h,-l	Xtriage
F_o, F_c correlation	0.90	EDS
Total number of atoms	12153	wwPDB-VP
Average B, all atoms $(Å^2)$	52.0	wwPDB-VP

Xtriage's analysis on translational NCS is as follows: The largest off-origin peak in the Patterson function is 19.72% 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: Z8O, $\mathrm{SO4}$

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	Mol Chain		lengths	Bond angles	
	Unam	RMSZ	# Z > 5	RMSZ	# Z > 5
1	А	0.45	0/2991	0.60	0/4077
1	В	0.42	0/3079	0.58	0/4181
1	С	0.42	0/3084	0.59	0/4190
1	D	0.43	0/3052	0.59	0/4141
All	All	0.43	0/12206	0.59	0/16589

There are no bond length outliers.

There are no bond angle outliers.

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	2919	0	2668	19	0
1	В	3004	0	2801	31	0
1	С	3008	0	2812	29	0
1	D	2978	0	2808	30	0
2	А	29	0	0	0	0
2	В	29	0	0	0	0
2	С	29	0	0	0	0
2	D	29	0	0	0	0
3	А	25	0	0	0	0



Mol	Chain	Non-H	H(model)	H(added)	Clashes	Symm-Clashes
3	В	15	0	0	1	0
3	С	30	0	0	1	0
3	D	15	0	0	0	0
4	А	12	0	0	0	0
4	В	12	0	0	0	0
4	С	10	0	0	0	0
4	D	9	0	0	1	0
All	All	12153	0	11089	91	0

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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 4.

The worst 5 of 91 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:35:ARG:NH1	1:D:35:ARG:HH12	1.59	1.01
1:C:35:ARG:HH12	1:D:35:ARG:NH1	1.63	0.95
1:A:251:VAL:HG13	1:A:257:ILE:HD13	1.53	0.88
1:C:35:ARG:HH12	1:D:35:ARG:HH12	0.81	0.75
1:B:330:CYS:O	1:B:334:THR:HG23	1.88	0.72

There are no symmetry-related clashes.

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	374/396~(94%)	365~(98%)	8 (2%)	1 (0%)	41	65
1	В	376/396~(95%)	362 (96%)	11 (3%)	3 (1%)	19	41
1	С	377/396~(95%)	366 (97%)	9 (2%)	2(0%)	29	53
1	D	369/396~(93%)	358~(97%)	11 (3%)	0	100	100



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Mol	Chain	Analysed	Favoured	Allowed	Outliers	Percentiles
All	All	1496/1584~(94%)	1451 (97%)	39~(3%)	6~(0%)	34 58

5 of 6 Ramachandran outliers are listed below:

Mol	Chain	Res	Type
1	В	100	ARG
1	В	104	GLY
1	С	268	ASP
1	А	388	GLU
1	С	265	GLN

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	Perce	ntiles
1	А	290/346~(84%)	282~(97%)	8(3%)	43	71
1	В	305/346~(88%)	299~(98%)	6(2%)	55	80
1	С	307/346~(89%)	297~(97%)	10 (3%)	38	66
1	D	306/346~(88%)	295~(96%)	11 (4%)	35	62
All	All	1208/1384~(87%)	1173 (97%)	35~(3%)	42	70

5 of 35 residues with a non-rotameric sidechain are listed below:

Mol	Chain	Res	Type
1	D	228	LEU
1	D	237	MET
1	D	287	MET
1	В	299	LEU
1	В	246	HIS

Sometimes side chains can be flipped to improve hydrogen bonding and reduce clashes. All (2) such side chains are listed below:



Mol	Chain	Res	Type
1	С	309	HIS
1	D	212	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)

21 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	Trung	Chain	Res	Link	Bo	ond leng	$_{\rm sths}$	В	ond ang	les
	Type	Chain	nes		Counts	RMSZ	# Z > 2	Counts	RMSZ	# Z > 2
3	SO4	А	503	-	4,4,4	0.11	0	$6,\!6,\!6$	0.42	0
3	SO4	А	504	-	4,4,4	0.17	0	6,6,6	0.25	0
3	SO4	В	503	-	4,4,4	0.18	0	$6,\!6,\!6$	0.30	0
3	SO4	С	503	-	4,4,4	0.25	0	6,6,6	0.43	0
3	SO4	С	507	-	4,4,4	0.22	0	6,6,6	0.19	0
3	SO4	В	502	-	4,4,4	0.78	0	$6,\!6,\!6$	0.15	0
3	SO4	С	505	-	4,4,4	0.18	0	6,6,6	0.27	0
3	SO4	В	504	-	4,4,4	0.16	0	$6,\!6,\!6$	0.23	0
3	SO4	С	506	-	4,4,4	0.16	0	6,6,6	0.11	0
2	Z8O	А	501	-	33,33,33	0.30	0	44,47,47	0.52	0
3	SO4	D	502	-	4,4,4	0.14	0	6,6,6	0.28	0
3	SO4	А	506	-	4,4,4	0.09	0	$6,\!6,\!6$	0.18	0
3	SO4	С	502	-	4,4,4	0.19	0	$6,\!6,\!6$	0.16	0



Mol	Turne	Chain	Dec	Res Link Bond lengths			Bond angles			
10101	Type	Unam	nes		Counts	RMSZ	# Z > 2	Counts	RMSZ	# Z >2
2	Z8O	С	501	-	33,33,33	0.32	0	44,47,47	0.63	0
3	SO4	А	505	-	4,4,4	0.24	0	$6,\!6,\!6$	0.27	0
3	SO4	D	504	-	4,4,4	0.20	0	6,6,6	0.10	0
3	SO4	D	503	-	4,4,4	0.21	0	6,6,6	0.40	0
3	SO4	С	504	-	4,4,4	0.13	0	6,6,6	0.19	0
3	SO4	А	502	-	4,4,4	0.17	0	$6,\!6,\!6$	0.11	0
2	Z8O	В	501	-	33,33,33	0.27	0	44,47,47	0.61	0
2	Z8O	D	501	-	33,33,33	0.32	0	44,47,47	0.65	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	Z8O	D	501	-	-	4/12/21/21	0/5/5/5
2	Z8O	А	501	-	-	3/12/21/21	0/5/5/5
2	Z8O	С	501	-	-	1/12/21/21	0/5/5/5
2	Z8O	В	501	-	-	1/12/21/21	0/5/5/5

There are no bond length outliers.

There are no bond angle outliers.

There are no chirality outliers.

5 of 9 torsion outliers are listed below:

Mol	Chain	Res	Type	Atoms
2	А	501	Z8O	C12-C10-N9-C8
2	D	501	Z8O	C12-C10-N9-C8
2	А	501	Z8O	O11-C10-N9-C8
2	D	501	Z8O	O11-C10-N9-C8
2	D	501	Z8O	C10-C12-C13-C14

There are no ring outliers.

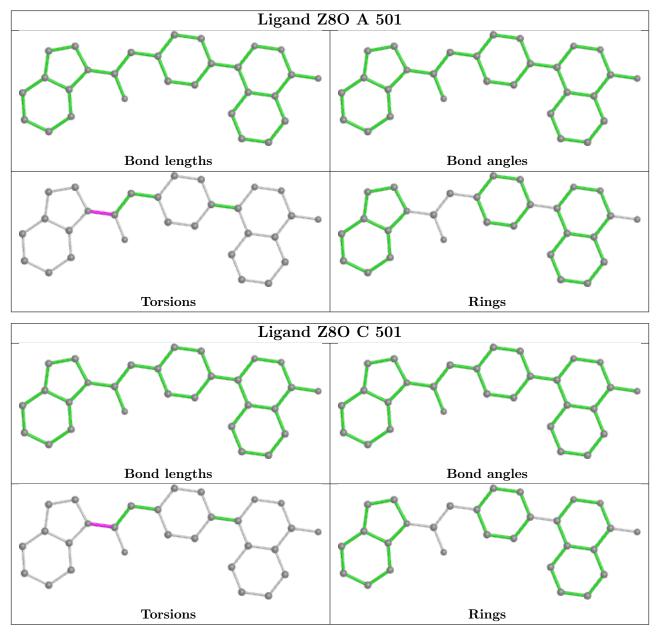
2 monomers are involved in 2 short contacts:

Mol	Chain	Res	Type	Clashes	Symm-Clashes
3	В	502	SO4	1	0
3	С	504	SO4	1	0

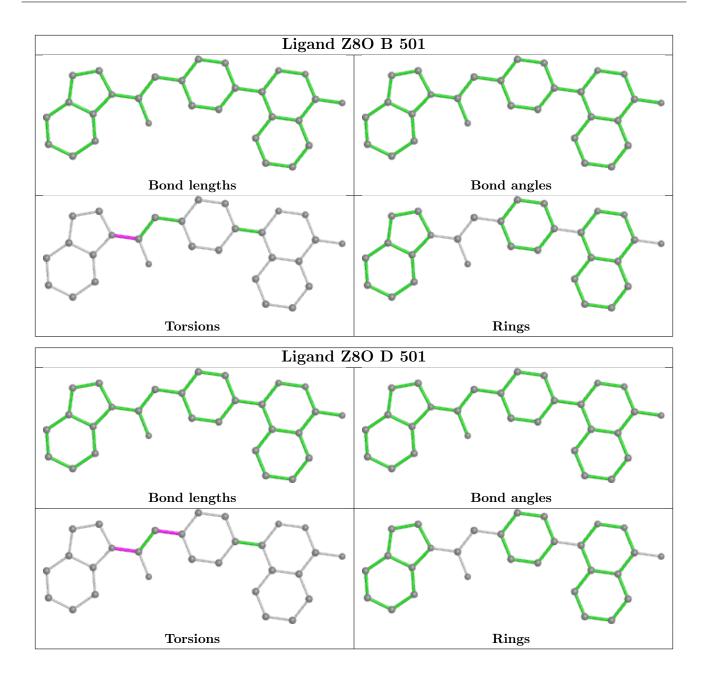
The following is a two-dimensional graphical depiction of Mogul quality analysis of bond lengths,



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	$\langle RSRZ \rangle$	# RSRZ > 2	$OWAB(Å^2)$	Q<0.9
1	А	384/396~(96%)	0.25	1 (0%) 94 95	29, 46, 70, 83	0
1	В	384/396~(96%)	0.37	9 (2%) 60 62	27, 51, 79, 95	0
1	С	385/396~(97%)	0.40	6 (1%) 72 74	32, 52, 77, 89	0
1	D	379/396~(95%)	0.35	6 (1%) 72 74	35, 55, 80, 88	0
All	All	1532/1584~(96%)	0.34	22 (1%) 75 77	27, 51, 77, 95	0

The worst 5 of 22 RSRZ outliers are listed below:

Mol	Chain	Res	Type	RSRZ
1	В	266	GLY	4.1
1	В	251	VAL	3.8
1	D	270	PHE	3.6
1	В	270	PHE	2.8
1	В	146	PHE	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.

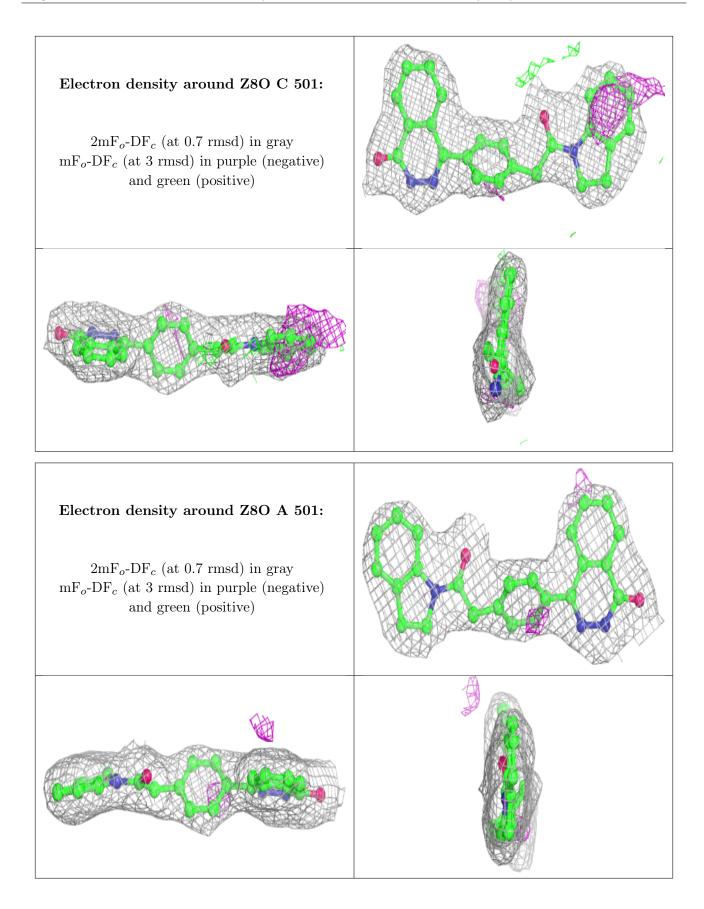


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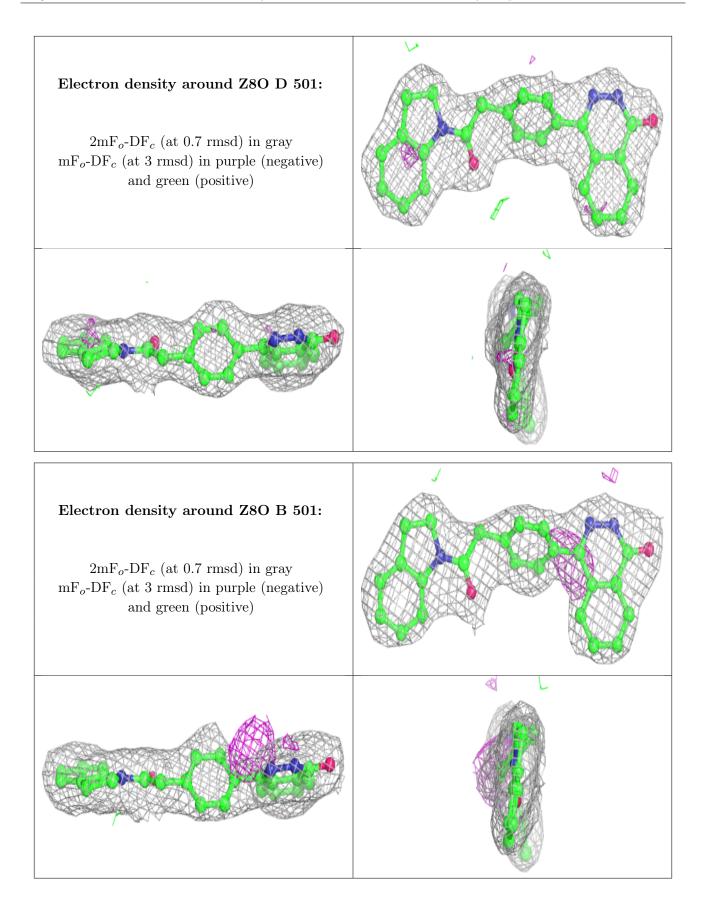
Mol	Type	Chain	Res	Atoms	RSCC	RSR	$\operatorname{B-factors}(\operatorname{\AA}^2)$	Q<0.9
3	SO4	С	506	5/5	0.54	0.26	150,150,151,151	0
3	SO4	С	502	5/5	0.88	0.21	125,125,125,125	0
3	SO4	А	505	5/5	0.89	0.17	106,107,107,107	0
3	SO4	С	505	5/5	0.90	0.15	103,103,103,103	0
3	SO4	А	506	5/5	0.90	0.19	107,107,107,107	0
3	SO4	А	502	5/5	0.91	0.17	123,123,123,123	0
3	SO4	С	503	5/5	0.91	0.19	81,81,81,81	0
3	SO4	В	504	5/5	0.94	0.13	97,97,97,97	0
3	SO4	С	507	5/5	0.95	0.16	97,97,97,97	0
3	SO4	А	503	5/5	0.96	0.12	70,70,70,71	0
2	Z8O	С	501	29/29	0.96	0.21	33,35,36,37	0
2	Z8O	А	501	29/29	0.96	0.21	36,39,42,42	0
3	SO4	D	502	5/5	0.96	0.14	90,91,91,91	0
2	Z8O	D	501	29/29	0.97	0.21	29,36,41,42	0
3	SO4	В	502	5/5	0.97	0.13	28,37,46,58	0
2	Z8O	В	501	29/29	0.97	0.23	32,33,34,35	0
3	SO4	D	504	5/5	0.97	0.10	106,106,106,106	0
3	SO4	С	504	5/5	0.98	0.15	79,80,80,80	0
3	SO4	В	503	5/5	0.98	0.15	66,66,66,66	0
3	SO4	D	503	5/5	0.98	0.15	$65,\!65,\!66,\!66$	0
3	SO4	А	504	5/5	0.98	0.15	60,60,60,60	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.

