wwPDB X-ray Structure Validation Summary Report (i)

Apr 21, 2024 – 11:41 pm BST

PDB ID	:	4ANW
Title	:	Complexes of PI3Kgamma with isoform selective inhibitors.
Authors	:	Foster, P.G.; Lougheed, J.C.
Deposited on	:	2012-03-22
Resolution	:	2.31 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 (i)) were used in the production of this report:

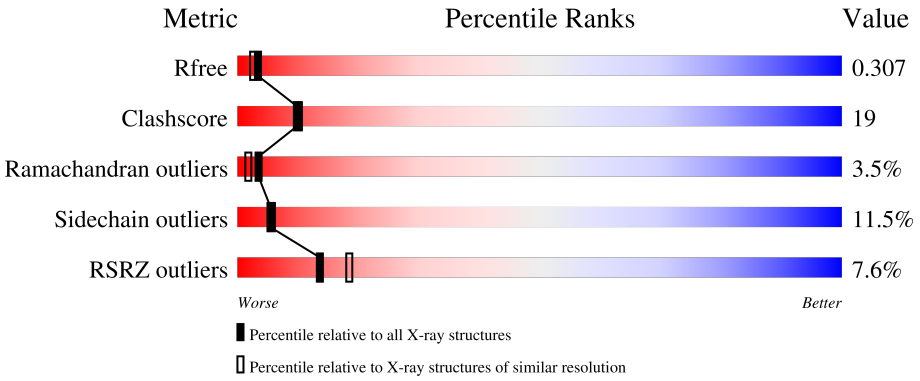
MolProbity	:	4.02b-467
Mogul	:	1.8.4, CSD as541be (2020)
Xtriage (Phenix)	:	1.13
EDS	:	2.36.2
buster-report	:	1.1.7 (2018)
Percentile statistics	:	20191225.v01 (using entries in the PDB archive December 25th 2019)
Refmac	:	5.8.0158
CCP4	:	7.0.044 (Gargrove)
Ideal geometry (proteins)	:	Engh & Huber (2001)
Ideal geometry (DNA, RNA)	:	Parkinson et al. (1996)
Validation Pipeline (wwPDB-VP)	:	2.36.2

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 2.31 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}	130704	5974 (2.34-2.30)
Clashscore	141614	6604 (2.34-2.30)
Ramachandran outliers	138981	6523 (2.34-2.30)
Sidechain outliers	138945	6523 (2.34-2.30)
RSRZ outliers	127900	5855 (2.34-2.30)

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 chain
1	A	980	6% 49% 29% 6% • 15%

2 Entry composition (i)

There are 4 unique types of molecules in this entry. The entry contains 6888 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.

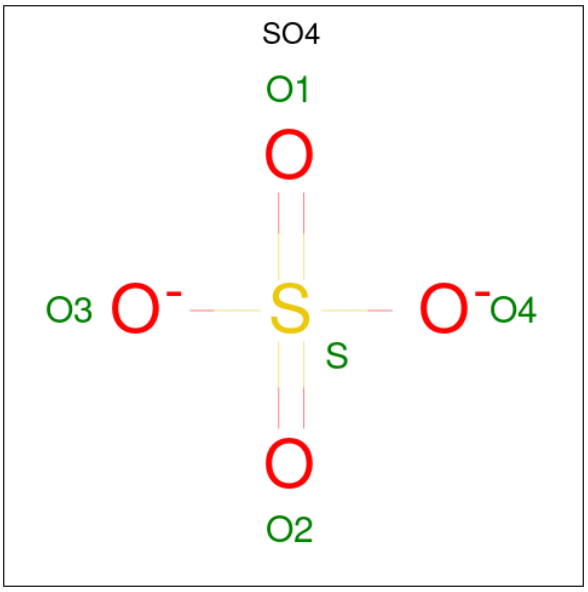
- Molecule 1 is a protein called PHOSPHATIDYLINOSITOL-4,5-BISPHOSPHATE 3-KINASE CATALYTIC SUBUNIT GAMMA ISOFORM.

Mol	Chain	Residues	Atoms					ZeroOcc	AltConf	Trace
			Total	C	N	O	S			
1	A	833	6757	4334	1153	1235	35	0	0	0

There are 21 discrepancies between the modelled and reference sequences:

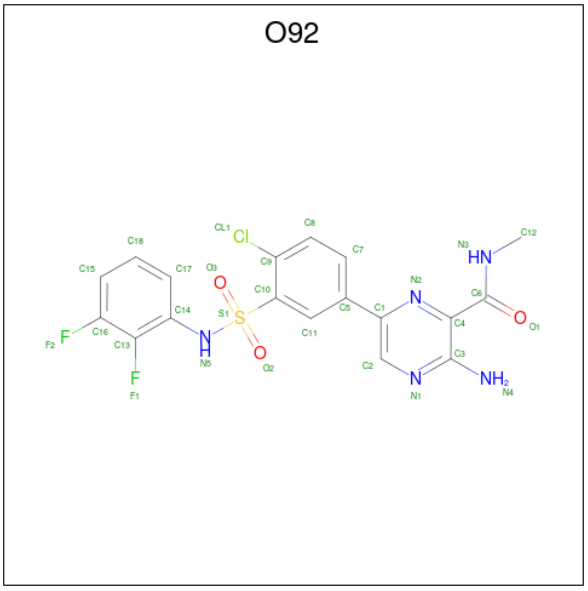
Chain	Residue	Modelled	Actual	Comment	Reference
A	139	MET	-	expression tag	UNP P48736
A	140	LEU	-	expression tag	UNP P48736
A	141	LEU	-	expression tag	UNP P48736
A	142	GLY	-	expression tag	UNP P48736
A	143	SER	-	expression tag	UNP P48736
A	1103	GLU	-	expression tag	UNP P48736
A	1104	PHE	-	expression tag	UNP P48736
A	1105	GLY	-	expression tag	UNP P48736
A	1106	LEU	-	expression tag	UNP P48736
A	1107	VAL	-	expression tag	UNP P48736
A	1108	PRO	-	expression tag	UNP P48736
A	1109	ARG	-	expression tag	UNP P48736
A	1110	GLY	-	expression tag	UNP P48736
A	1111	SER	-	expression tag	UNP P48736
A	1112	GLY	-	expression tag	UNP P48736
A	1113	HIS	-	expression tag	UNP P48736
A	1114	HIS	-	expression tag	UNP P48736
A	1115	HIS	-	expression tag	UNP P48736
A	1116	HIS	-	expression tag	UNP P48736
A	1117	HIS	-	expression tag	UNP P48736
A	1118	HIS	-	expression tag	UNP P48736

- Molecule 2 is SULFATE ION (three-letter code: SO4) (formula: O_4S).



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

- Molecule 3 is 3-AMINO-6-{4-CHLORO-3-[(2,3-DIFLUOROPHENYL)SULFAMOYL]PHENYL}-N-METHYLPYRAZINE-2-CARBOXAMIDE (three-letter code: O92) (formula: $C_{18}H_{14}ClF_2N_5O_3S$).



Mol	Chain	Residues	Atoms	ZeroOcc	AltConf
3	A	1	Total C Cl F N O S 60 36 2 4 10 6 2	0	1

- Molecule 4 is water.

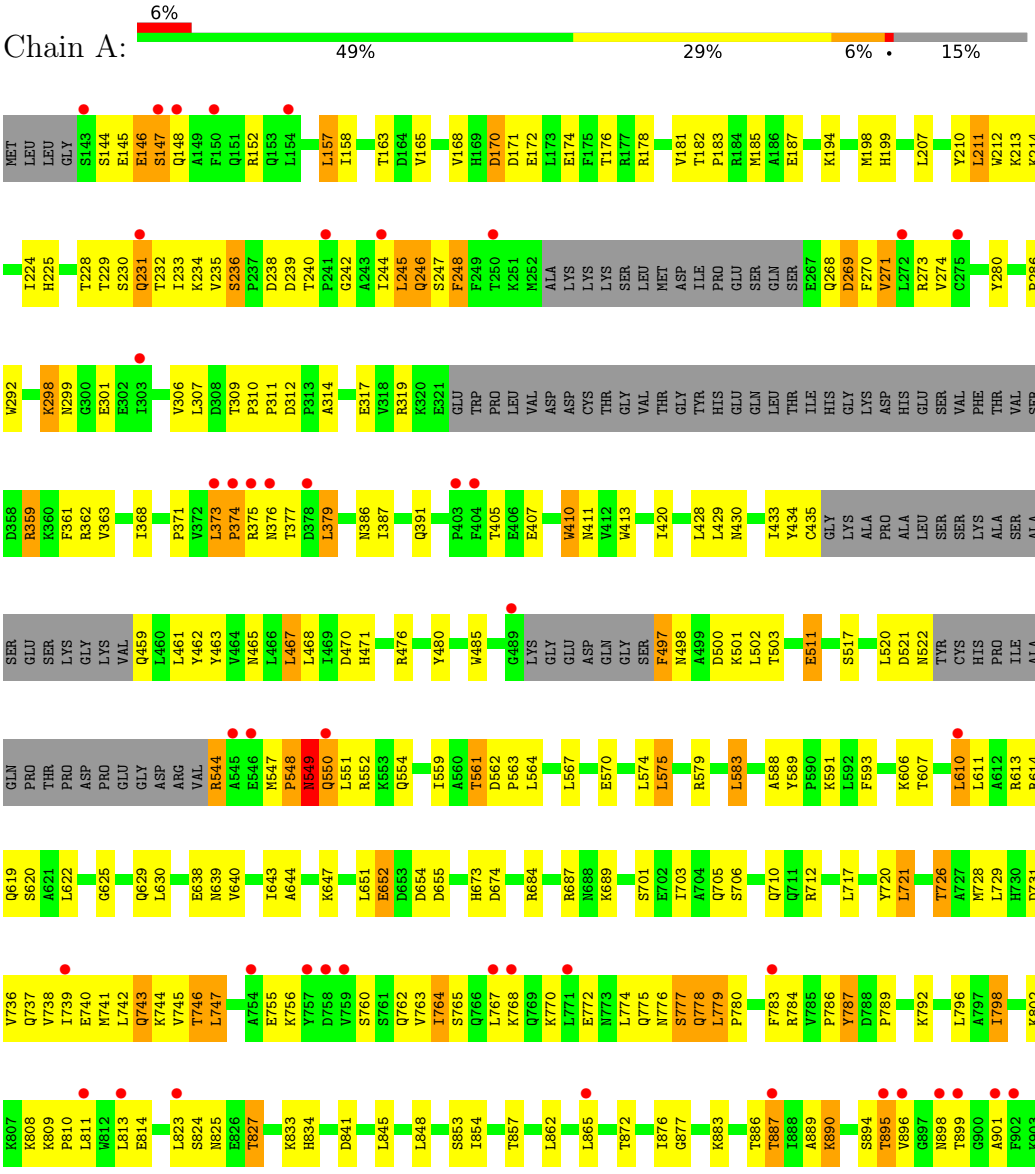
Mol	Chain	Residues	Atoms	ZeroOcc	AltConf
4	A	66	Total O 66 66	0	0

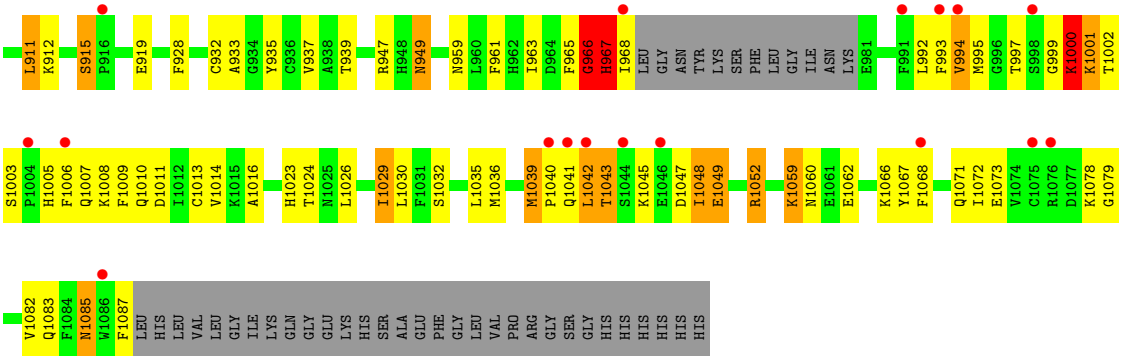
3 Residue-property plots (i)

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: PHOSPHATIDYLINOSITOL-4,5-BISPHOSPHATE 3-KINASE CATALYTIC SUBUNIT GAMMA ISOFORM

Chain A:




L911 K912 S915 P916 E919 F928 C932 A933 G934 Y935 C936 V937 A938 T939 R947 H948 N949 N959 L960 F961 H962 I963 D964 F965 G966 H967 I968 LEU GLY ASN TYR LYS SER PHE LEU GLY ILE ASN LYS E981 F991 L992 F993 V994 M995 G996 T997 S998 G999 K1000 K1001 T1002
S1003 P1004 H1005 F1006 Q1007 K1008 F1009 Q1010 D1011 I1012 C1013 V1014 K1015 A1016 H1023 T1024 N1025 L1026 I1029 L1030 F1031 S1032 L1035 M1036 M1039 P1040 Q1041 L1042 T1043 S1044 K1045 E1046 D1047 I1048 E1049 R1052 K1059 N1060 E1061 E1062 K1066 Y1067 F1068 Q1071 I1072 E1073 V1074 C1075 R1076 D1077 K1078 G1079
V1082 Q1083 F1084 N1085 W1086 F1087 LEU HIS LEU VAL LEU GLY ILE LYS GLN GLY GLU LYS HIS SER ALA GLU PHE GLY LEU VAL PRO ARG GLY SER GLY HIS HIS HIS HIS HIS HIS

4 Data and refinement statistics (i)

Property	Value	Source
Space group	C 1 2 1	Depositor
Cell constants a, b, c, α, β, γ	143.42Å 68.53Å 106.26Å 90.00° 95.26° 90.00°	Depositor
Resolution (Å)	71.43 – 2.31 71.41 – 2.31	Depositor EDS
% Data completeness (in resolution range)	99.2 (71.43-2.31) 98.3 (71.41-2.31)	Depositor EDS
R_{merge}	0.06	Depositor
R_{sym}	(Not available)	Depositor
$< I/\sigma(I) >$ [1]	1.43 (at 2.32Å)	Xtriage
Refinement program	REFMAC 5.2.0019	Depositor
R, R_{free}	0.235 , 0.296 0.246 , 0.307	Depositor DCC
R_{free} test set	2248 reflections (5.04%)	wwPDB-VP
Wilson B-factor (Å^2)	61.1	Xtriage
Anisotropy	0.139	Xtriage
Bulk solvent k_{sol}(e/Å^3), B_{sol}(Å^2)	0.35 , 58.9	EDS
L-test for twinning[2]	$< \|L\| > = 0.48$, $< L^2 > = 0.31$	Xtriage
Estimated twinning fraction	No twinning to report.	Xtriage
F_o,F_c correlation	0.94	EDS
Total number of atoms	6888	wwPDB-VP
Average B, all atoms (Å^2)	68.0	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.*

[1] Intensities estimated from amplitudes.

[2] 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.

5 Model quality

5.1 Standard geometry

Bond lengths and bond angles in the following residue types are not validated in this section: SO4, O92

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	0.89	2/6901 (0.0%)	0.92	11/9334 (0.1%)

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	A	0	3

All (2) bond length outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(Å)	Ideal(Å)
1	A	570	GLU	CB-CG	5.87	1.63	1.52
1	A	480	TYR	CG-CD2	-5.18	1.32	1.39

The worst 5 of 11 bond angle outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(^{o})	Ideal(^{o})
1	A	684	ARG	NE-CZ-NH1	12.36	126.48	120.30
1	A	684	ARG	NE-CZ-NH2	-7.88	116.36	120.30
1	A	575	LEU	CA-CB-CG	6.77	130.88	115.30
1	A	170	ASP	CB-CG-OD1	6.17	123.86	118.30
1	A	966	GLY	N-CA-C	5.98	128.04	113.10

There are no chirality outliers.

All (3) planarity outliers are listed below:

Mol	Chain	Res	Type	Group
1	A	1039	MET	Peptide

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Mol	Chain	Res	Type	Group
1	A	410	TRP	Peptide
1	A	965	PHE	Peptide

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	6757	0	6789	262	0
2	A	5	0	0	0	0
3	A	60	0	28	4	0
4	A	66	0	0	1	0
All	All	6888	0	6817	262	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 19.

The worst 5 of 262 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:777:SER:CB	1:A:778:GLN:HB2	1.59	1.31
1:A:967:HIS:H	1:A:968:ILE:HA	0.99	1.15
1:A:777:SER:HB3	1:A:778:GLN:CB	1.76	1.14
1:A:966:GLY:HA3	1:A:967:HIS:HB2	1.31	1.08
1:A:967:HIS:N	1:A:968:ILE:HA	1.61	1.02

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	Percentiles
1	A	819/980 (84%)	720 (88%)	70 (8%)	29 (4%)	3 2

5 of 29 Ramachandran outliers are listed below:

Mol	Chain	Res	Type
1	A	146	GLU
1	A	147	SER
1	A	211	LEU
1	A	374	PRO
1	A	778	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	Percentiles
1	A	749/874 (86%)	663 (88%)	86 (12%)	5 6

5 of 86 residues with a non-rotameric sidechain are listed below:

Mol	Chain	Res	Type
1	A	845	LEU
1	A	949	ASN
1	A	887	THR
1	A	905	GLU
1	A	1024	THR

Sometimes sidechains can be flipped to improve hydrogen bonding and reduce clashes. 5 of 28 such sidechains are listed below:

Mol	Chain	Res	Type
1	A	646	GLN
1	A	1083	GLN
1	A	743	GLN
1	A	949	ASN
1	A	710	GLN

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)

3 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	$\#\|Z\| > 2$
2	SO4	A	1188	-	4,4,4	0.33	0	6,6,6	0.75	0
3	O92	A	1189[B]	-	32,32,32	2.47	8 (25%)	43,47,47	2.11	8 (18%)
3	O92	A	1189[A]	-	32,32,32	2.48	8 (25%)	43,47,47	2.00	8 (18%)

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	O92	A	1189[A]	-	-	7/21/21/21	0/3/3/3
3	O92	A	1189[B]	-	-	2/21/21/21	0/3/3/3

The worst 5 of 16 bond length outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(Å)	Ideal(Å)
3	A	1189[B]	O92	O2-S1	8.02	1.52	1.43
3	A	1189[A]	O92	O2-S1	7.64	1.52	1.43
3	A	1189[A]	O92	C5-C1	-5.47	1.40	1.48
3	A	1189[B]	O92	C5-C1	-5.34	1.40	1.48
3	A	1189[B]	O92	O3-S1	5.33	1.49	1.43

The worst 5 of 16 bond angle outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(^{o})	Ideal(^{o})
3	A	1189[B]	O92	O3-S1-O2	-10.45	106.70	119.55
3	A	1189[A]	O92	O3-S1-O2	-9.16	108.29	119.55
3	A	1189[A]	O92	C15-C18-C17	-2.97	116.03	120.25
3	A	1189[B]	O92	O3-S1-N5	2.82	113.79	106.73
3	A	1189[B]	O92	O3-S1-C10	2.64	112.00	107.66

There are no chirality outliers.

5 of 9 torsion outliers are listed below:

Mol	Chain	Res	Type	Atoms
3	A	1189[A]	O92	C9-C10-S1-N5
3	A	1189[A]	O92	C17-C14-N5-S1
3	A	1189[A]	O92	C13-C14-N5-S1
3	A	1189[B]	O92	C9-C10-S1-N5
3	A	1189[B]	O92	C11-C10-S1-N5

There are no ring outliers.

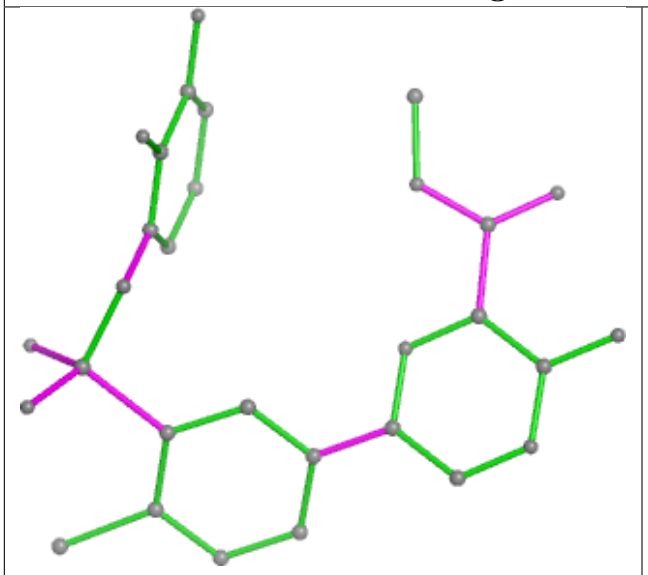
2 monomers are involved in 4 short contacts:

Mol	Chain	Res	Type	Clashes	Symm-Clashes
3	A	1189[B]	O92	1	0
3	A	1189[A]	O92	3	0

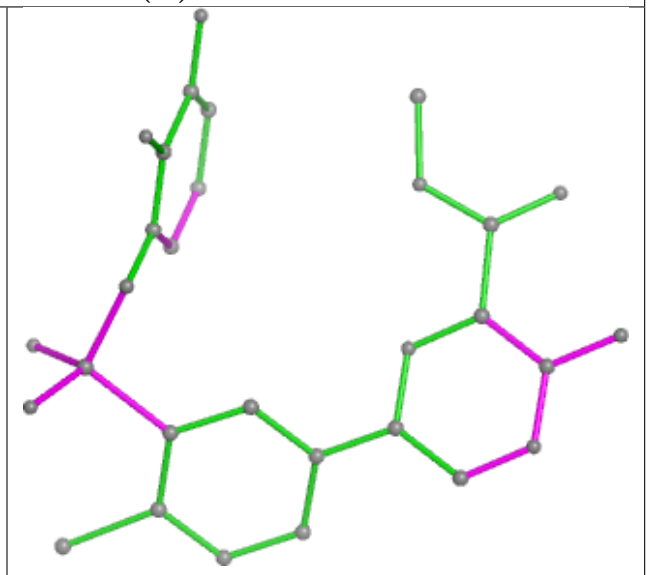
The following is a two-dimensional graphical depiction of Mogul quality analysis of bond lengths, bond angles, torsion angles, and ring geometry for all instances of the Ligand of Interest. In addition, ligands with molecular weight > 250 and outliers as shown on the validation Tables will also be included.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

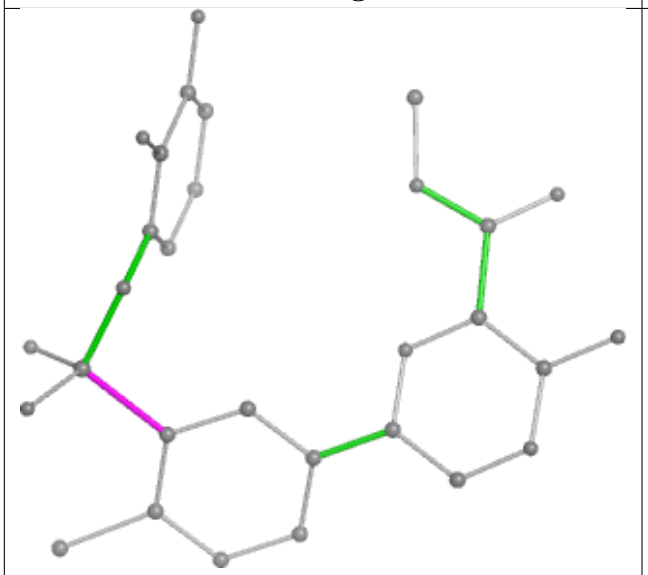
Ligand O92 A 1189 (B)



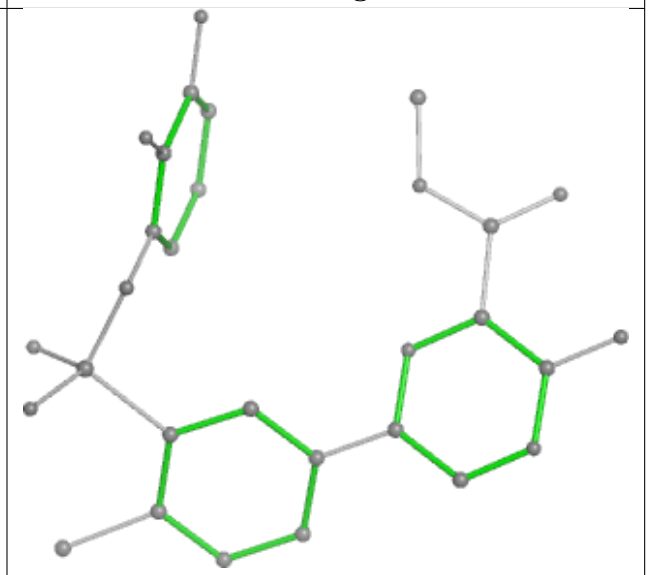
Bond lengths



Bond angles

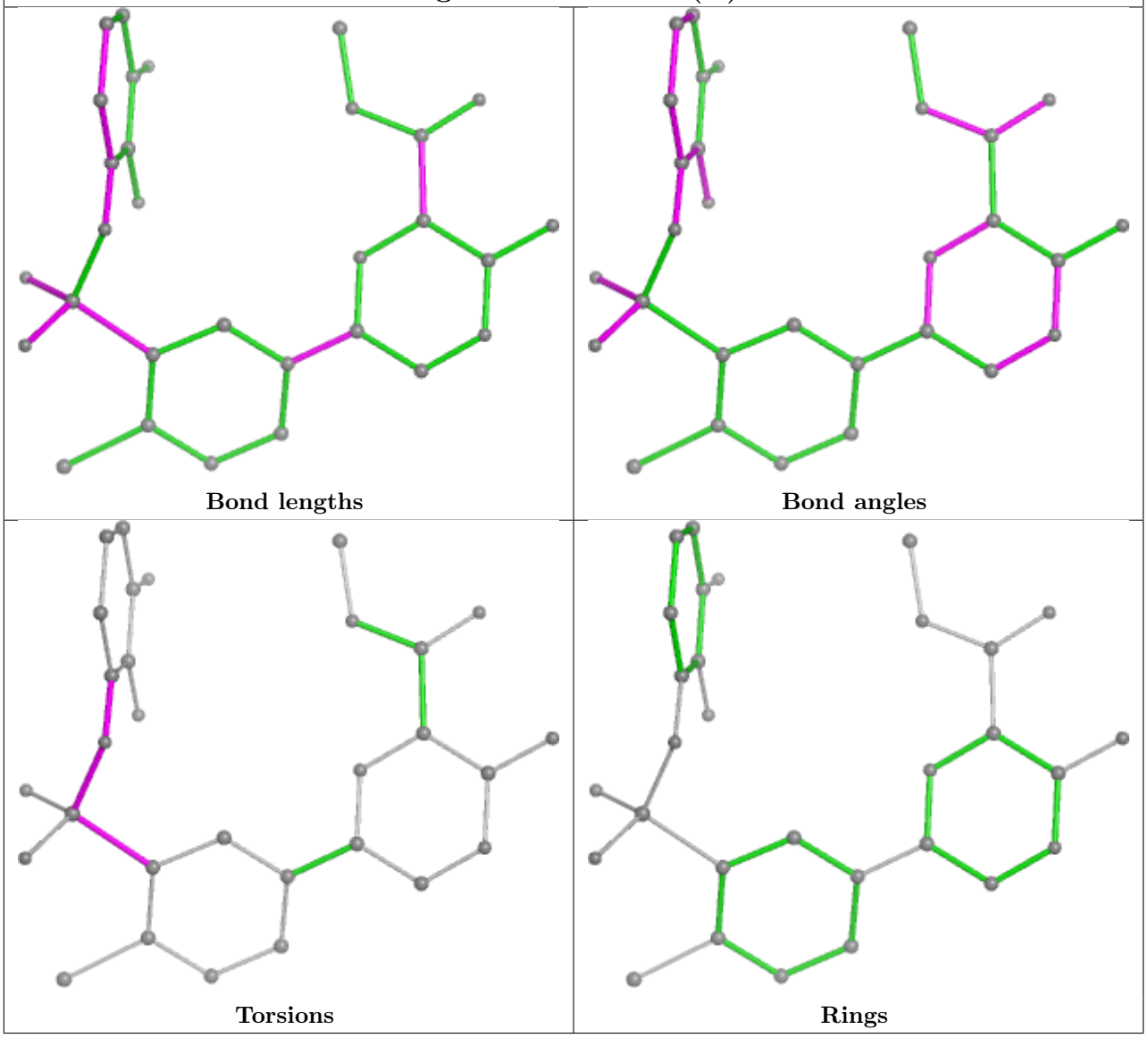


Torsions



Rings

Ligand O92 A 1189 (A)



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

6.1 Protein, DNA and RNA chains

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(Å^2)	Q<0.9
1	A	833/980 (85%)	0.63	63 (7%)	13	18	36, 65, 102, 115	0

The worst 5 of 63 RSRZ outliers are listed below:

Mol	Chain	Res	Type	RSRZ
1	A	1044	SER	8.9
1	A	757	TYR	7.9
1	A	998	SER	5.3
1	A	1086	TRP	5.2
1	A	895	THR	4.9

6.2 Non-standard residues in protein, DNA, RNA chains

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

6.3 Carbohydrates

There are no monosaccharides in this entry.

6.4 Ligands

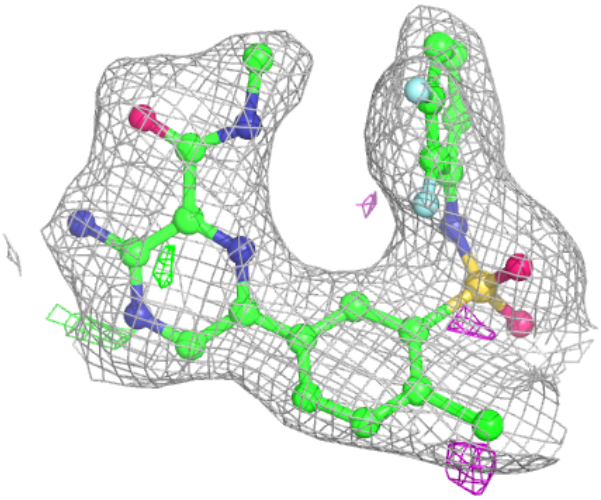
In the following table, the Atoms column lists the number of modelled atoms in the group and the number defined in the chemical component dictionary. The B-factors column lists the minimum, median, 95^{th} percentile and maximum values of B factors of atoms in the group. The column labelled 'Q< 0.9'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

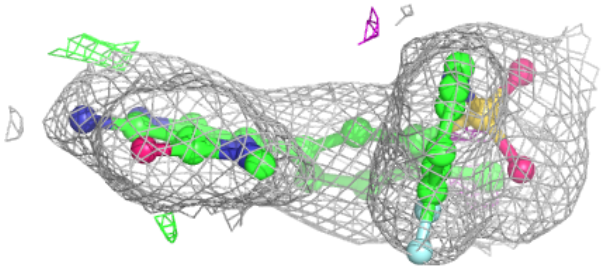
Mol	Type	Chain	Res	Atoms	RSCC	RSR	B-factors(Å^2)	Q<0.9
2	SO4	A	1188	5/5	0.94	0.12	75,76,79,81	0
3	O92	A	1189[A]	30/30	0.94	0.15	54,59,74,75	30
3	O92	A	1189[B]	30/30	0.94	0.15	50,57,71,71	30

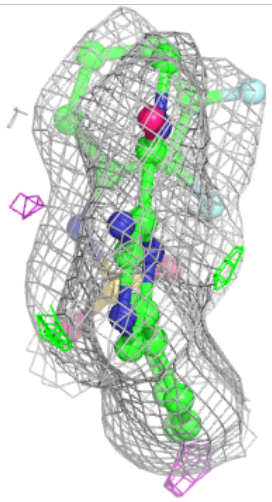
The following is a graphical depiction of the model fit to experimental electron density of all instances of the Ligand of Interest. In addition, ligands with molecular weight > 250 and outliers as shown on the geometry validation Tables will also be included. Each fit is shown from different orientation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

Electron density around O92 A 1189 (A):

$2mF_o$-DF_c (at 0.7 rmsd) in gray
mF_o-DF_c (at 3 rmsd) in purple (negative) and green (positive)

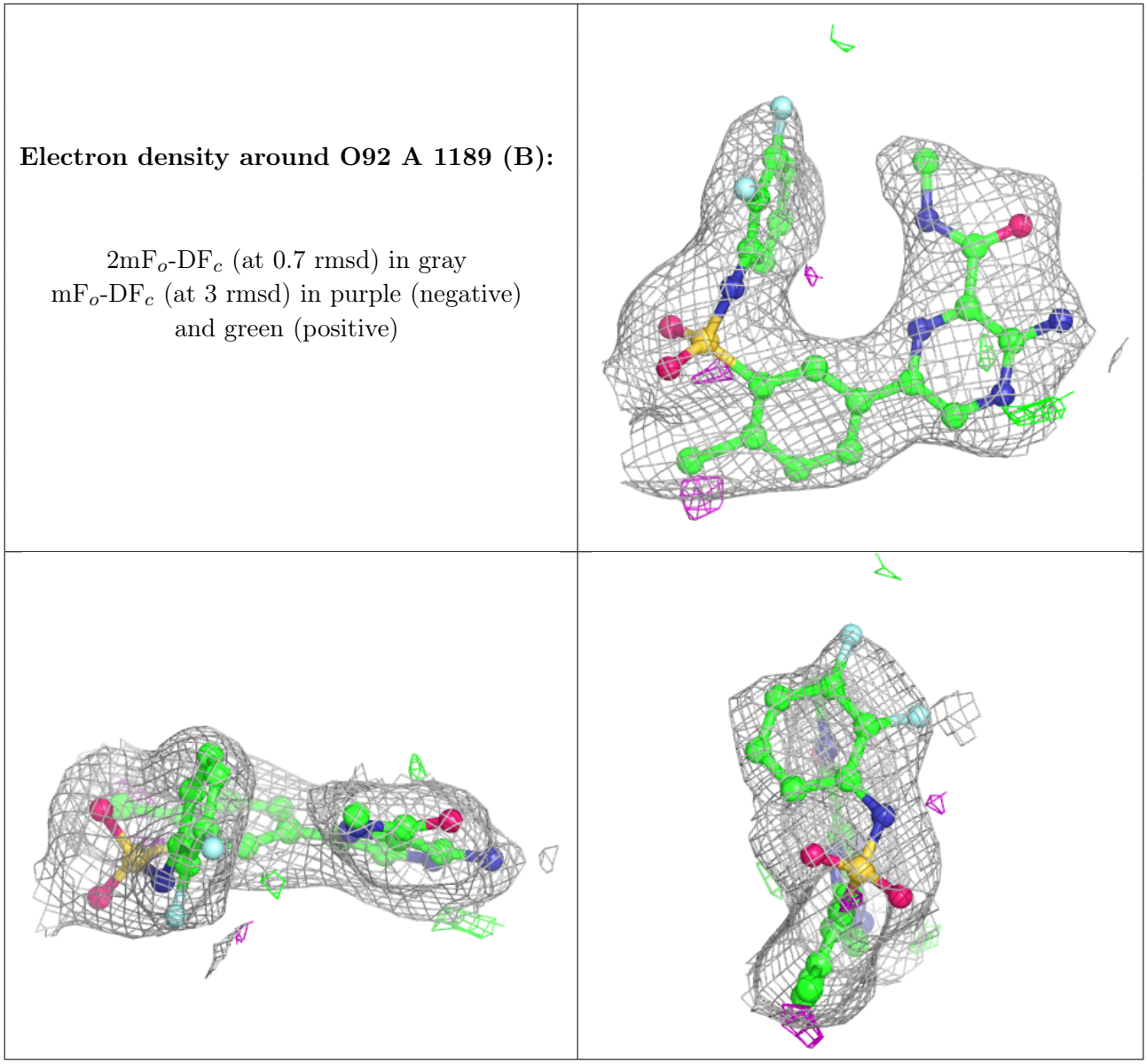






Electron density around O92 A 1189 (B):

$2mF_o$-DF_c (at 0.7 rmsd) in gray
mF_o-DF_c (at 3 rmsd) in purple (negative) and green (positive)



6.5 Other polymers (i)

There are no such residues in this entry.