



# Full wwPDB X-ray Structure Validation Report ⓘ

Mar 12, 2018 – 01:24 PM EDT

PDB ID : 5T31  
Title : Exploiting an Asp-Glu switch in Glycogen Synthase Kinase 3 to design paralog selective inhibitors for use in acute myeloid leukemia  
Authors : Stein, A.J.; Holson, E.B.; Wagner, F.F.; Cambell, A.J.  
Deposited on : 2016-08-24  
Resolution : 2.85 Å(reported)

This is a Full wwPDB X-ray Structure Validation 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 : rb-20030736  
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) : rb-20030736

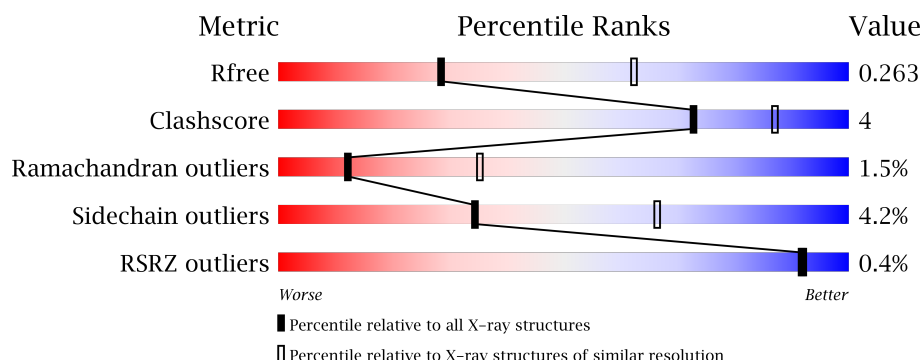
# 1 Overall quality at a glance

The following experimental techniques were used to determine the structure:

## *X-RAY DIFFRACTION*

The reported resolution of this entry is 2.85 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	2469 (2.90-2.82)
Clashscore	112137	2749 (2.90-2.82)
Ramachandran outliers	110173	2687 (2.90-2.82)
Sidechain outliers	110143	2690 (2.90-2.82)
RSRZ outliers	101464	2487 (2.90-2.82)

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	420	
1	B	420	

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
2	75F	B	501	-	-	-	X

## 2 Entry composition [i](#)

There are 2 unique types of molecules in this entry. The entry contains 5203 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.

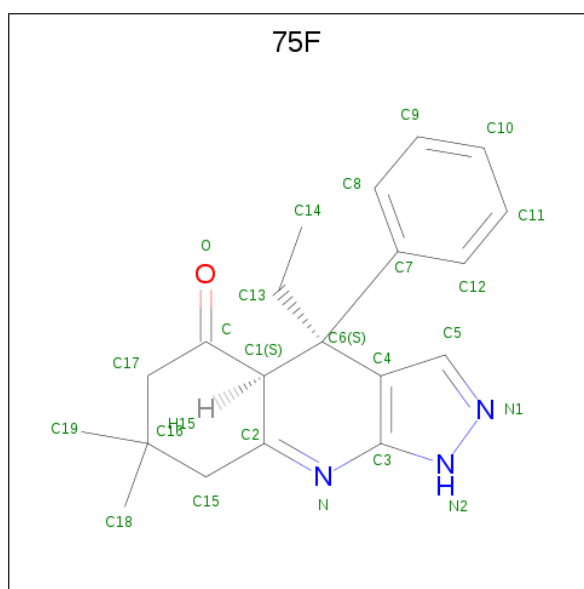
- Molecule 1 is a protein called Glycogen synthase kinase-3 beta.

Mol	Chain	Residues	Atoms						ZeroOcc	AltConf	Trace
1	A	335	Total	C	N	O	P	S	0	0	0
			2592	1666	432	482	1	11			
1	B	334	Total	C	N	O	P	S	0	0	0
			2563	1647	427	477	1	11			

There are 2 discrepancies between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	Comment	Reference
A	133	GLU	ASP	engineered mutation	UNP P49841
B	133	GLU	ASP	engineered mutation	UNP P49841

- Molecule 2 is (4S,4aS)-4-ethyl-7,7-dimethyl-4-phenyl-1,4,4a,6,7,8-hexahydro-5H-pyrazolo[3,4-b]quinolin-5-one (three-letter code: 75F) (formula: C<sub>20</sub>H<sub>23</sub>N<sub>3</sub>O).



Mol	Chain	Residues	Atoms				ZeroOcc	AltConf
2	A	1	Total	C	N	O	0	0
			24	20	3	1		

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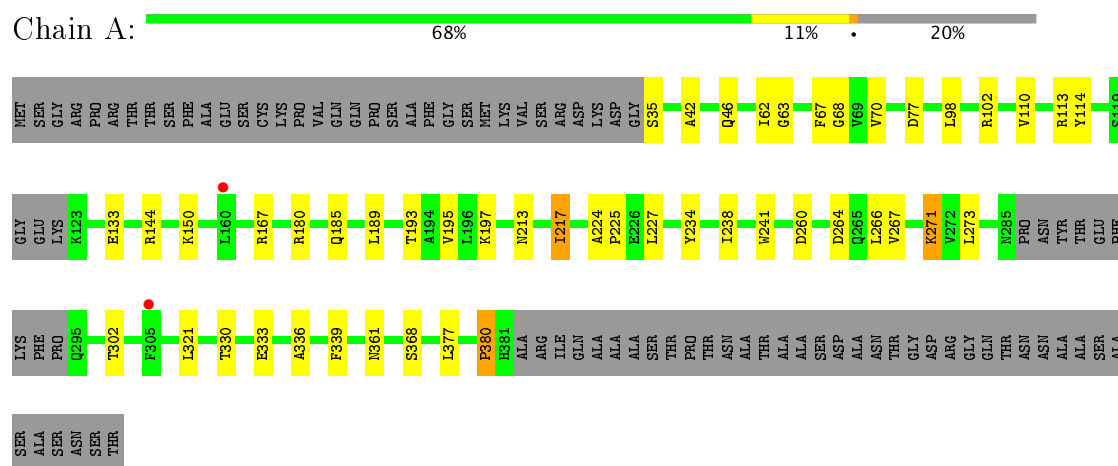
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Mol	Chain	Residues	Atoms				ZeroOcc	AltConf
2	B	1	Total	C	N	O	0	0
			24	20	3	1		

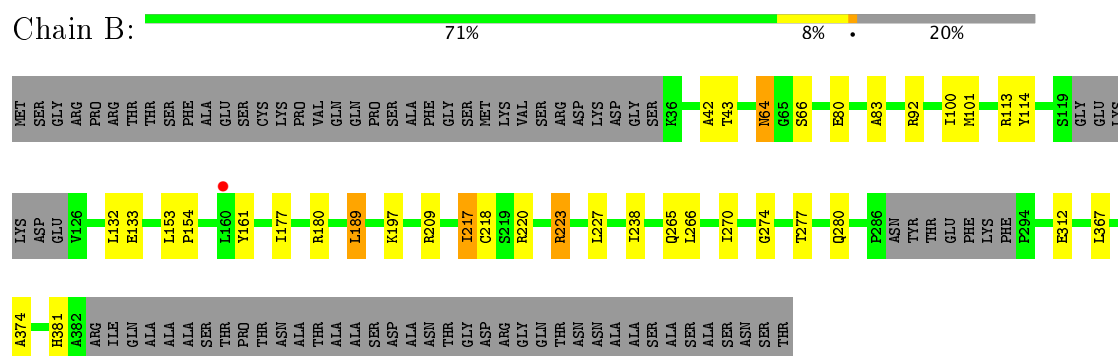
### 3 Residue-property plots

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: Glycogen synthase kinase-3 beta



- Molecule 1: Glycogen synthase kinase-3 beta



## 4 Data and refinement statistics

Property	Value	Source
Space group	C 1 2 1	Depositor
Cell constants a, b, c, $\alpha$ , $\beta$ , $\gamma$	107.96 Å   84.22 Å   117.22 Å 90.00°   94.69°   90.00°	Depositor
Resolution (Å)	47.41 – 2.85 47.41 – 2.85	Depositor EDS
% Data completeness (in resolution range)	97.0 (47.41-2.85) 97.0 (47.41-2.85)	Depositor EDS
$R_{merge}$	(Not available)	Depositor
$R_{sym}$	(Not available)	Depositor
$\langle I/\sigma(I) \rangle$ <sup>1</sup>	2.24 (at 2.86 Å)	Xtriage
Refinement program	REFMAC 5.8.0135	Depositor
R, $R_{free}$	0.196   ,   0.268 0.198   ,   0.263	Depositor DCC
$R_{free}$ test set	1206 reflections (5.32%)	DCC
Wilson B-factor (Å <sup>2</sup> )	80.8	Xtriage
Anisotropy	0.083	Xtriage
Bulk solvent $k_{sol}$ (e/Å <sup>3</sup> ), $B_{sol}$ (Å <sup>2</sup> )	0.30 , 45.0	EDS
L-test for twinning <sup>2</sup>	$\langle  L  \rangle = 0.49$ , $\langle L^2 \rangle = 0.32$	Xtriage
Estimated twinning fraction	No twinning to report.	Xtriage
$F_o, F_c$ correlation	0.94	EDS
Total number of atoms	5203	wwPDB-VP
Average B, all atoms (Å <sup>2</sup> )	82.0	wwPDB-VP

Xtriage's analysis on translational NCS is as follows: *The largest off-origin peak in the Patterson function is 4.85% 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: 75F, PTR

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	0.62	0/2636	0.80	0/3601
1	B	0.58	0/2609	0.77	1/3571 (0.0%)
All	All	0.60	0/5245	0.79	1/7172 (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	223	ARG	NE-CZ-NH2	-5.06	117.77	120.30

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	2592	0	2536	21	0
1	B	2563	0	2483	18	0
2	A	24	0	0	1	0
2	B	24	0	0	0	0
All	All	5203	0	5019	40	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 4.



All (40) 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:133:GLU:OE1	1:A:197:LYS:NZ	2.00	0.94
1:B:133:GLU:OE1	1:B:197:LYS:NZ	2.03	0.91
1:B:133:GLU:CD	1:B:197:LYS:NZ	2.34	0.80
1:B:133:GLU:OE2	1:B:197:LYS:NZ	2.15	0.79
1:A:133:GLU:OE2	1:A:197:LYS:NZ	2.17	0.78
1:A:133:GLU:CD	1:A:197:LYS:NZ	2.37	0.77
1:A:133:GLU:CD	1:A:197:LYS:HZ2	1.91	0.73
1:A:217:ILE:CD1	1:A:227:LEU:HD13	2.30	0.61
1:B:80:GLU:OE1	1:B:113:ARG:NH2	2.38	0.56
1:A:193:THR:HB	1:A:195:VAL:HG23	1.91	0.52
1:B:161:TYR:CZ	1:B:189:LEU:HD11	2.44	0.52
1:A:217:ILE:HD12	1:A:227:LEU:HD13	1.93	0.50
1:B:277:THR:O	1:B:280:GLN:N	2.45	0.48
1:B:42:ALA:HB1	1:B:114:TYR:HB3	1.95	0.48
1:B:133:GLU:CD	1:B:197:LYS:HZ3	2.00	0.48
1:B:180:ARG:HG2	1:B:238:ILE:HD11	1.96	0.48
1:B:270:ILE:O	1:B:274:GLY:N	2.44	0.48
2:A:501:75F:C12	2:A:501:75F:C	2.90	0.47
1:A:225:PRO:HD3	1:A:241:TRP:CE2	2.50	0.46
1:A:62:ILE:O	1:A:70:VAL:O	2.34	0.46
1:A:180:ARG:HG2	1:A:238:ILE:HD11	1.98	0.46
1:B:217:ILE:HD12	1:B:227:LEU:HD13	1.97	0.45
1:A:185:GLN:N	1:A:185:GLN:OE1	2.43	0.45
1:A:42:ALA:HB1	1:A:114:TYR:HB3	1.99	0.44
1:A:336:ALA:HB2	1:A:377:LEU:HD21	2.00	0.43
1:A:267:VAL:O	1:A:271:LYS:HG3	2.19	0.43
1:B:220:ARG:HG2	1:B:265:GLN:CG	2.47	0.43
1:A:98:LEU:O	1:A:102:ARG:HG3	2.20	0.42
1:A:224:ALA:HA	1:A:241:TRP:CD1	2.55	0.42
1:B:153:LEU:HD12	1:B:154:PRO:HD2	2.02	0.42
1:A:330:THR:OG1	1:A:333:GLU:HG3	2.20	0.42
1:B:367:LEU:HD22	1:B:374:ALA:HB2	2.01	0.42
1:B:217:ILE:O	1:B:223:ARG:HD3	2.20	0.42
1:B:100:ILE:HG23	1:B:177:ILE:HD13	2.02	0.42
1:A:227:LEU:HD21	1:A:234:TYR:CD2	2.56	0.41
1:A:264:ASP:O	1:A:267:VAL:HG12	2.21	0.41
1:A:273:LEU:HD11	1:A:321:LEU:O	2.21	0.40
1:B:83:ALA:HB3	1:B:132:LEU:HB2	2.03	0.40
1:A:67:PHE:CD1	1:A:68:GLY:N	2.89	0.40
1:B:64:ASN:HD22	1:B:64:ASN:HA	1.78	0.40

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	328/420 (78%)	291 (89%)	30 (9%)	7 (2%)	8	27
1	B	327/420 (78%)	304 (93%)	20 (6%)	3 (1%)	20	49
All	All	655/840 (78%)	595 (91%)	50 (8%)	10 (2%)	12	35

All (10) Ramachandran outliers are listed below:

Mol	Chain	Res	Type
1	A	150	LYS
1	A	380	PRO
1	B	92	ARG
1	A	63	GLY
1	A	368	SER
1	B	381	HIS
1	A	271	LYS
1	A	361	ASN
1	A	339	PHE
1	B	218	CYS

### 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	279/363 (77%)	265 (95%)	14 (5%)	28	59

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Mol	Chain	Analysed	Rotameric	Outliers	Percentiles	
1	B	273/363 (75%)	264 (97%)	9 (3%)	43	74
All	All	552/726 (76%)	529 (96%)	23 (4%)	34	66

All (23) residues with a non-rotameric sidechain are listed below:

Mol	Chain	Res	Type
1	A	35	SER
1	A	46	GLN
1	A	77	ASP
1	A	110	VAL
1	A	113	ARG
1	A	144	ARG
1	A	167	ARG
1	A	189	LEU
1	A	213	ASN
1	A	217	ILE
1	A	260	ASP
1	A	266	LEU
1	A	302	THR
1	A	380	PRO
1	B	43	THR
1	B	64	ASN
1	B	66	SER
1	B	101	MET
1	B	189	LEU
1	B	209	ARG
1	B	217	ILE
1	B	266	LEU
1	B	312	GLU

Some sidechains can be flipped to improve hydrogen bonding and reduce clashes. All (5) such sidechains are listed below:

Mol	Chain	Res	Type
1	A	108	ASN
1	A	265	GLN
1	A	365	GLN
1	B	64	ASN
1	B	365	GLN

### 5.3.3 RNA ⓘ

There are no RNA molecules in this entry.

## 5.4 Non-standard residues in protein, DNA, RNA chains ⓘ

2 non-standard protein/DNA/RNA residues 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$
1	PTR	A	216	1	15,16,17	0.75	0	19,22,24	1.09	1 (5%)
1	PTR	B	216	1	15,16,17	0.69	0	19,22,24	1.23	1 (5%)

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
1	PTR	A	216	1	-	0/9/11/13	0/1/1/1
1	PTR	B	216	1	-	0/9/11/13	0/1/1/1

There are no bond length outliers.

All (2) bond angle outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(°)	Ideal(°)
1	A	216	PTR	O3P-P-O2P	2.86	119.16	107.61
1	B	216	PTR	O3P-P-O2P	2.89	119.29	107.61

There are no chirality outliers.

There are no torsion outliers.

There are no ring outliers.

No monomer is involved in short contacts.

## 5.5 Carbohydrates

There are no carbohydrates in this entry.

## 5.6 Ligand geometry

2 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	75F	A	501	-	23,27,27	1.52	4 (17%)	21,42,42	1.79	4 (19%)
2	75F	B	501	-	23,27,27	1.59	4 (17%)	21,42,42	2.14	5 (23%)

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	75F	A	501	-	-	0/9/43/43	0/4/4/4
2	75F	B	501	-	-	0/9/43/43	0/4/4/4

All (8) bond length outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(Å)	Ideal(Å)
2	B	501	75F	C6-C4	-4.23	1.49	1.53
2	A	501	75F	C6-C4	-4.04	1.49	1.53
2	B	501	75F	C13-C6	-3.14	1.51	1.55
2	A	501	75F	C13-C6	-3.05	1.51	1.55
2	A	501	75F	C6-C7	-2.59	1.49	1.53
2	B	501	75F	C1-C	-2.51	1.48	1.52
2	B	501	75F	C6-C7	-2.30	1.49	1.53
2	A	501	75F	C1-C	-2.26	1.48	1.52

All (9) bond angle outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(°)	Ideal(°)
2	B	501	75F	C7-C6-C4	-7.54	102.00	111.58
2	A	501	75F	C7-C6-C4	-6.17	103.73	111.58
2	B	501	75F	C6-C1-C	-2.91	111.01	114.95
2	A	501	75F	C18-C16-C15	-2.48	105.89	109.83
2	B	501	75F	O-C-C1	-2.44	118.19	121.82
2	B	501	75F	C14-C13-C6	-2.33	111.23	114.85
2	A	501	75F	C15-C2-C1	2.51	121.78	115.62
2	A	501	75F	C15-C16-C17	3.13	113.42	108.15
2	B	501	75F	C15-C2-C1	3.23	123.54	115.62

There are no chirality outliers.

There are no torsion outliers.

There are no ring outliers.

1 monomer is involved in 1 short contact:

Mol	Chain	Res	Type	Clashes	Symm-Clashes
2	A	501	75F	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	334/420 (79%)	-0.22	2 (0%) 89 88	52, 79, 113, 139	0
1	B	333/420 (79%)	-0.17	1 (0%) 93 93	58, 81, 112, 125	0
All	All	667/840 (79%)	-0.19	3 (0%) 92 92	52, 80, 112, 139	0

All (3) RSRZ outliers are listed below:

Mol	Chain	Res	Type	RSRZ
1	A	305	PHE	2.4
1	A	160	LEU	2.3
1	B	160	LEU	2.1

### 6.2 Non-standard residues in protein, DNA, RNA chains [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
1	PTR	A	216	16/17	0.97	0.14	-	61,65,74,74	0
1	PTR	B	216	16/17	0.97	0.14	-	61,68,78,82	0

### 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( $\text{\AA}^2$ )	Q<0.9
2	75F	B	501	24/24	0.97	0.27	2.07	60,68,72,79	0
2	75F	A	501	24/24	0.97	0.24	1.59	61,70,73,74	0

## 6.5 Other polymers [i](#)

There are no such residues in this entry.